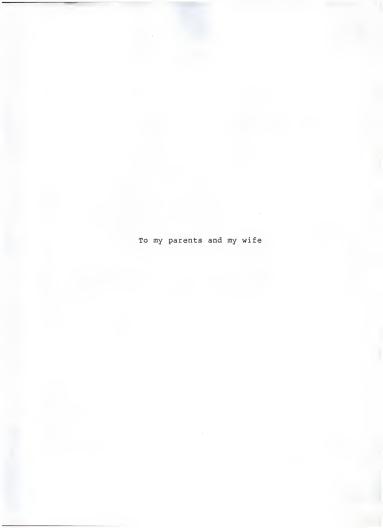
MASS PRODUCTION OF THE PREDACEOUS MITE, MACROCHELES MUSCAEDOMESTICAE (SCOPOLI)(ACARINA: MACROCHELIDAE), AND ITS POTENTIAL USE AS A BIOLOGICAL CONTROL AGENT OF HOUSE FLY, MUSCA DOMESTICA L. (DIPTERA: MUSCIDAE)

Ву

CHYI-CHEN HO

A DISSERTATION PRESENTED TO THE GRADUATE SCHOOL OF THE UNIVERSITY OF FLORIDA IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF DOCTOR OF PHILOSOPHY

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Abstract of Dissertation Presented to the Graduate School the University of Florida in Partial Fulfillment of the Requirements for the Degree of Doctor of Philosophy

MASS PRODUCTION OF THE PREDACEOUS MITE, MACROCHELES MUSCAEDOMESTICAE (SCOPOLI) (ACARINA: MACROCHELIDAE), AND ITS POTENTIAL USE AS A BIOLOGICAL CONTROL AGENT OF HOUSE FLY, MUSCA DOMESTICA L. (DIPTERA: MUSCIDAE)

Ву

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Major Department: Entomology and Nematology

This research has indicated that the predaceous macrochelid mite, Macrocheles muscaedomesticae

(Scopoli), is the key predator of house flies in caged-hen manure in Florida. This mite was found distributed in the same moisture level of manure as the house fly eggs and larvae while the other predators found in Florida are more abundant in manure with a drier moisture level. The population of this mite species is negatively correlated to the house fly larval population. Laboratory studies demonstrated that in their lifetime the adult female and male mites consume approximately 247 and 34 house fly eggs, respectively.

A study on the toxicity of 9 pesticides to adult female $\underline{\mathsf{M.}}$ muscaedomesticae showed that the mite strain studied may have developed resistance to coumaphos,

dichlorvos, dimethoate, malathion, and stirofos. This mite species showed a high potential for developing pesticide resistance with its short life cycle and high reproductive rate.

A mass-production method was developed for this mite species. Using spent house fly media, frozen house fly eggs, and a nematode, Protorhabditis sp., 2500 mites could be produced in 8 days from 34.5 female mites at 30 C. The nematode, Protorhabditis sp., which is found in Florida poultry manure, and its relationship to the rearing of these mites are discussed. Under these conditions, the generation time of this mite was 3.225 days and the mite population could increase as high as 69.85 times per generation. The possible ways to improve the mass-production method were discussed. Attempts were made to create a model to predict the harvest time and failed because of the complicated interactions between this mite species and the environment.

In laboratory tests, adult female mites in a ratio of 1/5, 1/10, and 1/20 to house fly eggs caused a mortality of 99.7%, 92.3%, and 77.7%, respectively.

The proper release time of this mite species in a IPM program to control house flies in poultry farms was discussed and a possible scouting method suggested.

CHAPTER 1

The Florida poultry industry is now nationally ranked as 8th in egg production and 12th in broiler production and contributes 5 to 7 percent of the total cash from Florida farm marketing. This value has ranged in dollars in the last decade from \$156.3 million in 1974 to \$233.1 million in 1983 (Anonymous 1983a). The Florida poultry business has been projected to maintain its current size and/or to expand in the '80s (Anonymous 1983b).

Florida producers, especially those with caged operations, face major problems with waste management and fly control (Koehler et al. 1979). This is brought into sharper focus when one considers that a laying hen will deposit wet droppings that are equal to the weight of feed it consumes (Axtell 1981) and that one white leghorn laying hen can excrete 0.25-0.40 pounds per day (Morgan et al. 1970). This means that a poultry unit with 50,000 birds could generate 5.6-9.0 tons of chicken manure per day, which could be exploited by the numerous species of filth flies.

In Florida, the most important and troublesome filth fly species in poultry farms is the house fly,

Musca domestica L. (Hogsette 1979, Koehler et al. 1979). Chapter 386 of Florida Statutes regulates excessive fly breeding and odors. Complaints by three responsible citizens about fly nuisance or excreta odors can result in a sanitary inspection of the suspected farm by the Department of Health and Rehabilitative Services (HRS). The inspected farm may eventually be closed if it fails to improve its sanitation and meet the HRS standards. In addition, the specks on the eggshells lower the market value of eggs, and extra labor is needed to wash and clean them. In 1983, approximately \$6 million was spent by poultry farmers for control of house fly problems (Butler, personal communications).

Numerous pesticides have been developed for house fly control. Many have been discontinued either because of the development of pesticide resistance or because of EPA findings that the compounds are carcinogenic or environmentally hazardous or both. This has brought about serious consideration of pest-management tactics other than chemical control.

The macrochelid mite, <u>Macrocheles muscaedomesticae</u> (Scopoli), is commonly found in cattle and chicken manure. It will travel on adult house flies and feed on house fly eggs and lst-instar larvae. It has been considered to be promising for house fly control. The research in this dissertation studied the mass

production of <u>M. muscaedomesticae</u>, the mite's annual dynamics in one poultry farm in Florida, the toxicity of nine insecticides currently in use for pest control in poultry farms, and a field evaluation of this mite as a control agent on the house fly. The ultimate goal of this study is to incorporate this mite with other control agents into an integrated program for managing the house fly on poultry farms.

CHAPTER 2

LITERATURE REVIEW

The common nuisance flies found at poultry farms in Florida are the house fly (Musca domestica L.), lesser house flies (Fannia spp.), the soldier fly (Hermetia illuscens (L.)), black garbage flies (Ophyra spp.), the stable fly (Stomoxys calcitrans (L.)), eye gnats (Chloropidae), hump backed flies (Phoridae), filter flies (Psychodidae), bottle flies (Calliphora spp., Cynomyopsis spp., Phaenicia spp.), and flesh flies (Sarcophaga spp.). The house fly is usually the most troublesome species (Hogsette 1979, Koehler et al. 1979). It costs millions of dollars to the Florida poultry industry every year to control house flies. The lesser house flies are important in cooler states, e.g. California (Dunning et al. 1978), but not in Florida. The soldier fly is not considered by most authors to be an economically important pest, although its larvae can be a serious nuisance on the floor and the walkways of the poultry house; and the larvae keep the manure liquified and break down the manure cones. Still, Bradley and Sheppard (1984) considered it a competitive agent that limits the production of other flies. The other species of flies are not a major problem at most

poultry farms in Florida due to their normal low density and are only a serious problem in special cases (Koehler et al. 1979).

The House Fly

The house fly, Musca domestica (L.), is a cosmopolitan pest, infamous for its transmitting both human and animal diseases. Harwood and James (1979) pointed out that behaviorally and morphologically, the house fly is remarkably effective in transmitting disease. Behaviorally, it is synanthropic; it freely frequents human food, animal (including human) excrement, and garbage, and it constantly emits vomit spots for liquifying solid materials for digestion. Morphologically, its pseudotracheal system of proboscis and hairy body, especially the legs and proboscis, are excellent for collecting contaminants. Greenberg (1971) listed the organisms associated with the house fly in a 146-page publication. These organisms included viruses, bacteria, rickettsia, fungi, protozoa, cestodes, nematodes, arachnids (pseudoscorpions, spiders, mites), myriapods, insects, gastropods, and a few vertebrates. Unfortunately, their relationship was not listed.

House fly adults oviposit in poultry and animal manure, waste feed, garbage, decaying vegetation, and in almost any kind of warm, moist organic materials (Harwood and James 1979, Koehler et al. 1979, Hogsette

1979). The adult can fly as far as 32 km, and may disperse 5-6 km in large numbers. Usually they disperse 1-3 km (Harwood and James 1979). Koehler et al. (1979) gave similar data: the adult may migrate 1-4 miles, usually 1/4 to 2 miles. Greenberg (1971) reported the house fly had dispersed 11.8 km within 24 hours.

House flies have certain behavior patterns around poultry houses. Anderson (1964) found in studies at poultry farms in California that 85% of the house flies were found within the poultry house. Visual counts at night in the poultry house showed that almost the entire indoor population rested on or near the ceiling and remained there for 12 to 16 hours. Anderson also observed that the greatest fly activity on the droppings occurred from early to mid-afternoons. In Florida, Koehler et al. (1979) stated the house fly will remain outside until about 1200 or 1300 in the hot summer and then move into the poultry house. At night and on cold rainy days they stay inside the poultry house.

The development time of the house fly from egg to adult is greatly influenced by temperature. Harwood and James (1979) gave as representative figures, 45 days at 16 C, 27 days at 18 C, 20 days at 20 C, 16 days at 25 C, and 10 days at 30 C. In Florida, a house fly can complete its life cycle in 6-10 days. The average life span of the house fly is approximately 30 days. A

typical female house fly can lay up to 1300 eggs in her lifetime. Eggs are normally laid in clusters of 75-100 eggs, preferably in new animal waste or moist organic matter. The egg usually hatches within 12 to 24 hours. During the warm weather there can be 2 or more generations per month depending on the media. The warm humid weather of Florida permits the house fly to breed year-round (Koehler et al. 1979). For the mass production of house flies in an insectary under 28.3 $^{\pm}$ 2 C, at 70% $^{\pm}$ 5% R.H., it takes 8-12 hours for the egg stage, 6 days for larval stage, 4 days for pupal stage, making a life cycle of 10-11 days. With a preoviposition period of 3 days, and a 30-day life span, the average female is able to lay 1080 eggs in her lifetime (Morgan 1981c).

Since a hen can generate 0.25-0.40 pounds of wet droppings every day (Morgan et al. 1970), if this waste is improperly managed, a single hen can produce enough media for 200 fly larvae each day (Koehler et al. 1979). Therefore, a commercial chicken farm of 50,000 birds is capable of producing 10,000,000 flies a day! It is interesting to note that Morgan et al. (1970) tried to use house fly larvae to biodegrade and hence to manage poultry manure.

Natural Enemies of the House Fly

Greenberg (1971) published an extensive list of organisms associated with house flies, but he did not designate which were natural enemies of the fly. West (1951) does discuss the parasites, predators, symbionts, and commensals of the house fly.

Parasitoids

In Morgan's review of the research on the parsasitic wasps of house flies and other muscoid flies (1977, 1981c), he pointed out that the species most effective in attacking fly pupae were in the family Pteromalidae. Those with the potential for fly control are Spalangia endius Walker, S. cameroni Perkins, S. nigra Latreille, S. nigroaenea Curtis, S. muscidarum Richardson (=S. nigroaenea), S. afra Silvestri, S. fallax Masi, S. gemina sp. n., S. longepetiolata sp.n., S. melanogastra Masi., S. obscura sp.n., S. seyrigi Risbee, S. simplex Perkins, S. sulcifera sp.n., Muscidifurax raptor Girault and Sanders, M. zaraptor Kogan and Legner, M. raptorellus Kogan and Legner, M. raptoroides Kogan and Legner, M. uniraptor Kogan and Legner, Nasonia vitripennis (Walker), Mormoniella vitripennis (Walker) (=N. vitripennis (Walker)), Pachycrepoideus vindemiae (Rondani) (=P. dubius Ashmead), and Tachineaphagus zealandicus Ashmead.

In California, McCoy (1965) studied the control effect of Musca domestica and Fannia sp. on poultry farms by the mass liberation of Muscidifurax raptor. The percentage of parasitization of indigenous house fly pupae was on average less than 25 percent. This low percentage was due to M. raptor is inability to find house fly pupae in areas where their greatest concentration was located. The same conclusion was drawn by Morgan et al. (1981). Legner and Brydon (1966) studied the population of pupal parasites of the flies in southern California poultry ranches. Among the 6 active parasite species, M. raptor and Spalangia endius accounted for more than 95% of the observed parasitism on Fannia femoralis and Ophyra leucostoma. M. raptor was prominent in the cooler, more humid months while S. endius was prominent in the hotter and drier months. Legner and Dietrick (1972) reported a 6.5 times lower density of Fannia sp. and almost twice the percentage of parasitism with inundative release of M. raptor, S. endius, and Tachinaephagus zealandicus. Additional work of Legner and Dietrick (1974) demonstrated that the inoculative release of S. endius, M. raptor, M. zaraptor, and T. zealandicus over 20 months in southern California poultry ranches significantly reduced the population density of Musca domestica, Muscina stabulans, F. canicularis, F. bemoralis, O. leucostoma, S. calcitrans, and Phaenicia spp. Olton and Legner

(1975) inoculatively released $\underline{\mathbf{T}}$. $\underline{\mathbf{zealandicus}}$, $\underline{\mathbf{S}}$. $\underline{\mathbf{endius}}$, and $\underline{\mathbf{M}}$. $\underline{\mathbf{raptor}}$, after which 46% of house fly pupae were parasitized.

In North Carolina, Rutz and Axtell (1979) used a sustained release of \underline{M} . \underline{raptor} in caged-layer poultry houses and reported a twofold increase in both the rate of parasitism and the proportion of \underline{M} . \underline{raptor} in the parasitoid population. Release of the same parasite in broiler-breeder poultry houses produced a significant reduction in the house fly population (Rutz and Axtell 1981).

Sheppard and Kissam (1981) combined 2 control strategies on a poultry farm in Georgia by releasing $\underline{\mathbf{M}}$. $\underline{\mathbf{raptor}}$ and treating the house fly resting surface with diflubenzuron. On a season-long basis, the highest average percentage of parasitism was 29.4% and comprised mainly indigenous parasitoids, $\underline{\mathbf{Spalangia}}$ nigroaenea Curtis in particular.

In Florida, Mitchell et al. (1974) did a survey of the parasites of flies at poultry farms in north central Florida and found five species: Muscidifurax raptor, Aphaereta musebecki, Spalangia cameroni, S. endius, S. nigra. Butler et al. (1981) did a similar survey at poultry houses in north Florida using the pupal trap method and found S. endius (26%), S. nigraoaenea (36%), and S. cameroni (38%) to be the most common one. Pickens (1981) in Maryland failed to get

satisfactory control of the house fly in poultry coops by releases of the pupal parasitoid, Pachycrepoideus vindamiae. Morgan et al. (1975) released 445,200 S. endius females per week in a small caged-layer farm of 6700 birds. The house fly population was completely suppressed within 35 days. Further studies on S. endius and M. raptor by Morgan et al. (1981) found that M. raptor did not seek Musca domestica pupae beneath the soil surface and did not function well in dry, hot weather. The authors also noted that S. endius did not seem to be able to locate and parasitize house fly pupae effectively if the poultry manure was too wet.

Predators

Predators of the house fly found in poultry droppings include beetles and their larvae, dipterous larvae, earwigs, hunting wasps, ants, spiders, birds, and mites (Anderson 1965, Legner 1971).

Dipterous larvae

The larvae of the black garbage fly, Ophyra

leucostoma (Wied), is predaceous on the larvae of other
muscoid flies. It can kill as many as 20 fly larvae per
day. It often will kill more than it can eat (Anderson
1964, Anderson and Poorbaugh 1964). In the "index of
predation potential" calculated by Peck (1969), O.

<u>leucostoma</u> had the highest value, ≥ 226.4 . Another species, <u>O</u>. <u>capensis</u> (Weid) was the most abundant on a British poultry farm (Conway 1973).

It has been suggested that the false stable fly,

<u>Muscina stabulans</u> (Fallen), was a voracious predator of
other fly larvae (Anderson 1964). However, Anderson and
Poorbaugh (1964) reported that it was rarely predaceous
in their study.

Coleopterans

Hartman (1970) reported good management of poultry manure chiefly through the biocontrol of flies by the larvae and adult mealworm beetle of an unknown species. Peck and Anderson (1969) surveyed arthropod predators of fly larvae in poultry droppings in northern California. Three species each of Staphylinidae and Histeridae and larvae of O. leucostoma and M. stabulans were collected in addition to mites. Legner (1971) studied the ambient arthropod complex in poultry waste in southern California. He recorded Coleoptera (Anthicidae, Histeridae, Hydrophilidae, Monotomidae, Scarabaeidae, Staphylinidae), Dermaptera (Labiduridae), Hemiptera (Anthocoridae), Lepidoptera, Pseudoscorpionida, and Acarina (Cheyletidae, Saproglyphidae, Stigmaeidae, Uropodidae). By inoculating house fly eggs and with the insecticide check method, he also demonstrated that a mortality of

53.4-99.4% resulted from the presence of predatory and scavenger arthropods. Pfeiffer and Axtell (1980) studied the Coleoptera of caged-layer manure in North Carolina. They collected over 120 species belonging to 31 families. Anthicidae. Cucujideae. Dermestidae. Historidae, Hydrophilidae, Mycetophagidae, Nitidulidae, Ptilidae, Rhizophilidae, Scarabaeidae, Scolytidae, Staphylinidae, and Tenebrionidae were the common families. The histerid, Carcinops pumilio (Erichson), and the tenebrionid, Alphitobius disperinus (Panzer), were most abundant. Bills (1973) reported C. pumilio is the final dominant arthropod in the accumulating manure in the deep-pit poultry house in England. C. pumilio had a predation potential index in house fly control of 97.0 according to Peck (1969). Peck (1969) studied the predation of Philonthus politus, P. sordidus, Staphilus maxillosus villosus (staphylinids), and Margarinotus merdarius, C. pumilio (histerids). In the laboratory, they all preyed on immature stages of the house fly and coastal fly, Fannia femoralis Stein.

Predaceous mites

Three families of mites are known to prey upon fly eggs and/or larvae in poultry manure. They are Macrochelidae, Parasitidae, and Uropodidae, which all belong to the suborder Gamasida (=Mesostigmata)(Axtell 1963a, 1981; Conway 1973; Peck and Anderson 1969).

Parasitidae were most abundant, then Macrochelidae, and then Uropodidae. Parasitidae appeared earliest in the season, followed by Macrochelidae and Uropodidae. Their population declined in the same order (Axtell 1970a, Peck and Anderson 1969). Willis and Axtell (1968) studied the populations of Macrocheles muscaedomesticae and Fuscuropoda vegetans following the accumulation of manure. It showed F. vegetans invading and establishing after M. muscaedomesticae. Nevertheless, the results of Rodriguez et al. (1970) showed that the population of F. vegetans peaked twice a year, not necessarily following macrochelids each time.

Not a great deal is known about the parasitid mites associated with filth flies. The adults and deutonymphs of <u>Parasitus gregarius</u> Ito preferred feeding on house fly larvae over fly eggs. The deutonymphs of this species preferred the nematode, <u>Rhabditis elongata</u>, over the fly larvae and/or eggs (Ito 1977a). Ito (1977b) studied its mass production. With five deutonymphs supplied with <u>R. elongata</u> as food, 204.6, 318.2, and 389.2 mites, mostly deutonymphs, were harvested after 5, 10, and 15 days, respectively. The adults and deutonymphs of another parasitid mite, <u>Poecilochirus monospinosis</u>, commonly found in poultry and cattle manure, are predaceous on fly eggs and lst-instar larvae (Axtell 1981).

The most commonly encountered uropodid mite predaceous on filth flies is Fuscuropoda vegetans (Geer). It is predaceous on fly eggs and 1st-instar larvae. The male, female, and deutonymph ate 1.36, 1.66, and 1.34 house fly eggs per mite per day, respectively (Jalil and Rodriguez 1970b, O'Donnell and Axtell 1965). But Willis and Axtell (1968) found it could not penetrate the chorion of house fly eggs and fed only upon the 1st-instar larvae. It can feed, however, on the eggs of the little house fly, Fannia canicularis (O'Donnell and Nelson 1967). Female and male mites consumed 7.0 and 6.4 eggs per mite in 11 and 9 days, respectively. Willis and Axtell (1968) reared the mite on house fly eggs, nematodes, wheat germ, brewers yeast, and white bread. Peck and Anderson (1969) noticed that the mite stays in drier manure. It was found gregarious and deeper (2-4 cm) within the manure cone where 1st-instar fly larvae usually are present. It was dominant or nearly dominant after 5-6 weeks of manure accumulation. It moved slowly and was phoretic on dung beetles (Willis and Axtell 1968).

Jalil and Rodriguez (1970b) studied the biology and odor perception of \underline{F} . $\underline{vegetans}$. When reared in the laboratory at 27 C, 50-55% R.H. and on nematodes, this mite had a life cycle of 23 \pm 0.28 and 24 \pm 0.75 days for males and females, respectively. Females that were fed on nematodes had a higher fecundity than those fed on

house fly eggs (7 eggs/day vs 4 eggs/day). The preoviposition period and oviposition period averaged 8 and 24 days, respectively. The average life span of fertilized females (234 days) was longer than unfertilized females (169 days). The life span of males averaged 210 days. The most important structure for odor perception appeared to be the long apical setae on the tarsus of leg 1. Both adult and immature stages preferred nematodes to house fly eggs or larvae. Ito (1971) tested and found F. vegetans could feed on 5 species of nematodes. In studies on feeding preference, Ito reported both adults and deutonymphs of F. vegetans (=Uroobovella marginata Koch) preferred nematodes (Rhabditis elongata) to the house fly larvae or eggs (Ito 1977a), which is similar to the results of Jalil and Rodriguez (1970b). Ito (1977b) supplied nematodes every 5 days, and the mite population increased from 5 adults to 448.5 mostly immature individuals in 20 days. Thereafter, the population fluctuated and remained relatively constant. At the 50th day, there were 445.8 mites, mostly nymphs.

The control of the house fly by \underline{F} . $\underline{\text{vegetans}}$ was studied by O'Donnell and Axtell (1965), Peck (1969), Rodriguez et al. (1970), and Willis and Axtell (1968). In the study of Rodriguez et al. (1970), this mite produced 87% control on house fly eggs. In the other studies, the control was not greater than 56.5%.

The literature of as many as 16 dung-inhabiting macrochelid mite species was reviewed by this researcher. They were Macrocheles cristati Costa, M. eurygaster Kr., M. glaber (Muller), M. matrius (Hull), M. medarius (Berl.), M. muscaedomesticae (Scopoli), M. parapisentii Costa, M. penicilliger (Berl.), M. periculatus Berl., M. peregrinus Krantz, M. perglaber Filipponi and Pegazzano, M. robustulus (Berl.), M. saceri Costa, M. subbadius (Berl.), M. vernalis (Berl.), and Glyphtholaspis confusa (Foa).

 $\underline{\text{M. cristati}}$, $\underline{\text{M. parapisentii}}$, $\underline{\text{M. robustulus}}$, $\underline{\text{M. }}$. $\underline{\text{saceri}}$, $\underline{\text{M. medarius}}$, and $\underline{\text{M. muscaedomesticae}}$ were reported from Israel by Costa (1966, 1967). The biology of the first 4 species were studied.

M. peregrinus and M. eurygaster were collected in South Africa by Krantz (1981). The former was introduced into northern Australia to control the eggs and larvae of the buffalo fly, Haematobia irritans exigua De Meijere, and the bush fly, Musca vetustissima Walker, and has since established itself in Australia (Wallace and Holm 1983).

In Egypt, Abdel-Gawaad et al. (1976) reported a low house fly population coincided with abundant $\underline{\text{M}}$. glaber. At 30 C, when fed on house fly eggs, the developmental time (egg to adult) and longevity of M. matrius averaged 2.86 and 26.3 days for female mites and 2.5 and 19.3 days for male mites, respectively.

When fed on 1st-instar house fly larvae, the life cycle and longevity averaged 3.3 and 24.6 days for female mites and 2.7 and 16.1 days for male mites, respectively. During the immature and adult stages, the female mite consumed an average of 12.9 and 118.4 house fly eggs or 11.3 and 101.0 1st-instar larvae while the male mite consumed 11.6 and 76.4 house fly eggs or 9.6 and 69.8 1st-instar larvae, respectively. Feeding on house fly eggs increased the fecundity and prolonged the oviposition period of the mite. When fed with house fly eggs or larvae, the female laid on the average 63.8 and 40.7 eggs, respectively. Generation time (egg to egg) averaged 4.9 and 6.0 days, respectively (Soliman et al. 1978).

In Italy, Filipponi (1955) studied the natural association between M. muscaedomesticae and the house fly, Musca domestica. The number of species studied was then increased to 6 species by 1963, M. muscaedomesticae, M. glaber, M. peniculatus, M. perglaber, M. robustulus, and M. scutatus (Filipponi and di Delupis 1963). All species fed on house fly eggs and 1st-instar larvae and nematodes of the family Rhabditidae. The reproductive rate of these mites was low when fed on yeast, enchytraeids, and springtails. Differential preference to nematode and house fly eggs existed between species and between adults and nymphs of the same species. In the same year, Filipponi and

Francaviglia (1963) reported that the thelytokous M. peniculatus may lay eggs, or eggs which have a developed embryo, or larvae, or even protonymphs according to breeding conditions. Studies on the influence of food, temperature, crowding, ventilation, and substrate moisture showed that the food condition was the most important factor. Filipponi and Petrelli (1966) studied the capacity of population increase of the house fly. In 1967 Filipponi and Petrelli reported on the autoecology and the capacity of increase of M. muscaedomesticae under different temperatures, which was completed in 1971 (Filipponi et al. 1971). The mite's optimum reproduction temperature ranged from 28 C to 32 C. The highest r (intrinsic capacity of increase) value, 0.906, was obtained at 30 C. M. perglaber and M. robustulus were studied by Filipponi and Petrelli and Filipponi and Mosna (Cicolani 1979). Filipponi and Passariello (1969) again studied M. peniculatus on the influence of temperature on its fecundity, longevity, and speed of multiplication. By 1971 the influences of temperature on the population increase of 4 macrochelid mites, M. muscaedomesticae, M. penicilliger, M. peniculatus, and M. subbadius, were compared (Filipponi 1971). The optimum temperature ranges for them were 28-34 C, 22-26 C, 24-28 C, and 25-29 C, respectively. The highest r values were 0.906 at 30 C, 0.323 at 24 C, 0.564 at 26 C, and 0.865 at 29 C, respecitively. Another mite, \underline{M} . \underline{M} matrius was studied by Filipponi and Cicolani (1974). It is an arrhenotokous, oviparous species, but it would be viviparous under overcrowding or starvation conditions. Its r is greatest, 0.810, at 34 C. The optimum temperature ranged from 24 C to 34 C. \underline{M} . $\underline{Subbadius}$ was reported again in 1979 with 2 more temperatures studied, 31 and 33 C. It was found that the optimum temperature was 31 C with r = 0.878 instead of the 0.865 at 29 C. (Cicolani et al. 1977). The biology, ecology, and morphology of \underline{M} . $\underline{Vernalis}$ (Berl.) were reported by Cicolani in 1980.

In the United States, Axtell (1961a) collected M. robustulus, M. subbadius, M. medarius, M. muscaedomesticae, and G. confusa from cow and horse manure in New York state. Except for M. medarius, they were studied for their predation rate on house fly eggs and 1st-instar larvae. During a 3-day period, 9.9, 4.6, 2.7, and 1.2 house fly eggs and/or 1st-instar larvae were consumed by G. confusa, M. robustulus, M. muscaedomesticae, and M. subbadius respectively. Axtell (1961b) reported their potential as biocontrol agents of house flies. Axtell (1963a) added M. glaber and M. matrius to the list. In 211 manure samples of dairy cattle, horse, sheep, chicken, and duck manure collected from 92 farms, the Macrochelidae were the most abundant, followed by Uropodidae, Parasitidae,

Oribatidae, Laelaptidae, and 4 other families. Individually, M. muscaedomesticae was the most abundant and was usually the only macrochelid mite found in chicken manure.

Cicolani (1979) compared 7 macrochelid species that had been studied in Italy. Each species reached its maximum r at a different temperature. Six of them were higher than the house fly. M. muscaedomesticae had the highest, r = 0.906 at 30 C; followed by M. subbadius, 0.878 at 31 C; M. perglaver, 0.817 at 27 C; M. matrius, 0.810 at 34 C; M. robustulus, 0.706 at 29 C; M. peniculatus, 0.564 at 26 C; in contrast to the 0.370 of house fly at 28 C or M. penicilliger which had a r of 0.323 at 24 C.

Macrocheles muscaedomesticae (Scopoli, 1772)

Classification

Following the taxonomic system of Krantz (1978) this mite belongs to

Order

Acari

Suborder

Gamasida

Supercohort Monogynaspides

Cohort

Gamasina

Superfamily Eviphidoidea

Family Macrochelidae

Genus

Macrocheles

Species

muscaedomesticae

M. muscaedomesticae under different names has long been reported to be associated with flies. Pereira and de Castro (1945) reviewed the literature on macrochelids that travel on flies. They listed 37 synonyms of this mite and emended the current name, Macrocheles muscaedomesticae (Scopoli, 1772). Based on laboratory cultures that were fed house fly eggs, their report carefully described every stage of this mite. Krantz and Filipponi (1964) rectified the number of synonyms to 8.

Distribution

M. muscaedomesticae is a cosmopolitan species. It has been collected from Australia (Krantz and Filipponi 1964), Brazil (Pereira and de Castro 1945), Canada (Chant 1960), Czechoslovakia (Pereira and de Castro 1945), England (Krantz and Filipponi 1964), Iran (Rak 1972), Israel (Costa 1967), Italy (Filipponi 1955), Japan (Ito 1971), Mexico (Hoffmann et al. 1974), New Zealand (Emberson 1973, 1980), Saudi-Arabia (Samainak 1979), the USSR (Axtell 1964) and the United States (Ewing 1913).

In the United States, Ewing (1913) first reported it as a new parasite of the adult house fly. He named this mite Macrocheles muscae. Specimens were collected from Ithaca, New York, and Corvallis, Oregon. Since then it has been reported from Massachussetts (Steve

1959), Kentucky (Rodriguez and Wade 1961), California (Anderson 1964), North Carolina (Axtell 1964), and Florida (William and Rodgers 1976). Sanders and Dobson (1966) collected macrochelids from bovine manure in Indiana without the identifying.

Habitats

M. muscaedomesticae is generally found in livestock and poultry manure. It is found primarily in the outermost layer and near the peak of the manure cone where the fly eggs are usually accessible (Willis and Axtell 1968). However, it also has been collected from nest of the American kestrel, Falco sparverius in New York City (Philips and Norton 1979); nests of Porzana tabuensis plumbea, the spotless crake; Turdus merula, the black bird; and also from decayed bird carcasses in New Zealand (Emberson 1980).

Dispersal

Macrochelid mites are commonly phoretic on Diptera and Coleoptera. Only the adult female mites, frequently those not fertilized, attach (Pereira and de Castro 1947). Macrochelids that inhabit domestic dung piles tend to be phoretic on synanthropic flies. Those that inhabit pastoral dung pads tend to be phoretic on beetles (Krantz 1983).

M. muscaedomesticae has been found phoretic on house flies (Ewing 1913, Filipponi 1955, Hoffmann et al. 1974, Pereira and de Castro 1945, Rak 1972), Fannia canicularis (Axtell 1964, Hoffmann et al. 1974, Rodriguez et al. 1962, Steve 1959), Stomoxys calcitrans (Axtell 1964, Hoffmann et al. 1974, Williams and Rodgers 1976), Muscina stabulans (Emberson 1973, Hoffmann et al. 1974), Dermatobia hominis (Moya Borja 1981), Ophyra sp. (Hoffmann et al. 1974), Ophyra antrax and Eristalis tenax (Axtell 1964). Petrova reported 16 fly species, but the main phoretic hosts of M. muscaedomesticae are Musca domestica and O. leucostoma (Axtell 1964).

There was no preference as to sex of house fly for phoresy. The mite preferred the abdomen to the head, then the thorax and the venter to the dorsum of the house fly. Furthermore, the mites preferred to attach to the intersegmental membranes between main body regions. Temperature and density of the mites and the house flies also affect the rate of phores (Jalil and Rodriguez 1970a). Both sexes of the adult house fly possess a water soluble volatile chemical (or chemicals) that is highly attractive, and even more attractive than house fly eggs, to adult females of this mite (Jalil and Rodriguez 1970a, Wicht et al. 1971). Five- to 7-day-old flies had a larger amount of attractant than newly emerged flies. This chemical(s)

was apparently related to an amino derivative of a carbohydrate (Wicht et al. 1971). Farish reported that the phoretic behavior of this mite was correlated with the ovipositional behavior of the house fly. The house fly preferred to oviposit in fresh manure. M.

<u>muscaedomesticae</u> was attracted to fresh manure, and the attractiveness declined as the manure aged. Phoretic behavior took place when the manure attractiveness was lower than the flies and vice versa (Axtell 1969).

The palps and tarsi 1 were important sensory receptors in the act of phoresy and predation. The first tarsi were involved in odor reception in locating the fly, and contacting the fly with the palps was necessary for attachment (Farish and Axtell 1966).

When phoretic, the mite inserted its chelicerae into the intersegmental membrane or gripped a bristle of the house fly to secure itself (Filipponi 1955, Jalil and Rodriguez 1970a). In laboratory testing, when the mite attached to the house fly, the house fly lost weight and had a shorter life span (Jalil and Rodriguez 1970a, Kinn 1966). Ewing (1913) and Jalil and Rodriguez (1970a) thought the mite obtained nutrition from the house fly. On the other hand, Pereira and de Castro (1945) and later Krantz (1983) considered the relationship was phoretic only, with no feeding involved. However, Butler (1964), using dyes, believed that M. muscaedomesticae did feed on adult house flies.

Filipponi collected 724 M. muscaedomesticae from 67,389 house flies. Less than 1.1% of the house flies carried the mite (Axtell 1964). King (1964) reported only about 2% of the house flies in nature carry this mite. Yet studies of Axtell (1964) showed it varying in different cases.

King (1964) also reported that under cloudy weather conditions this mite stayed on the surface of the substrate longer than on sunny days. One thousand mites can disperse evenly to an area of 16-18 inches in radius in approximately 10 minutes. Tests showed it was photonegative when the light source was bright. Furthermore, it was more photonegative to direct light rather than to diffused light (the light on cloudy days).

This mite preferred to stay in 21.1-23.9 C at 10% or 50% relative humidity and 23.9-26.7 C at 80% or 90% relative humidity. The choice of 21.1-23.9 C at 50% R.H. was greater than at 10% R.H. The temperature preference seemed to change at around 50% R.H. These choices were different from the results of Singh et al. (1967). In their studies, this mite selected 16.7 $^{\pm}$ 0.9 C (=62.1 $^{\pm}$ 1.6 F) at the three studied R.H. levels of 45-50%, 80-90% and 90-100%. It also opted for the highest available humidity in all test choices. It performed well in discriminating the differences in humidity. Still, Rodriguez et al. (1962) reported the

population of this mite was most active in manure strata at 80 to 85 F. Materials containing 65% to 70% moisture were ideal for mite activity and movement.

Food Habits

M. muscaedomesticae was found to feed on house fly eggs and 1st-instar larvae (Axtell 1961a, Filipponi 1955, Pereira and de Castro 1945) and nematodes (Filipponi and di Delupis 1963, Ito 1971, Rodriguez et al. 1962, Singh and Rodriguez 1966). Adult mites preferred house fly eggs over 1st-instar larvae, and over nematodes (Filipponi 1955, O'Donnell and Axtell 1965, Rodriguez and Wade 1961, Rodriguez et al. 1970), while nymphs preferred nematodes over house fly eggs (Filipponi and di Delupis 1963, Ito 1977a, Rodriguez et al. 1962). Over 8 species of nematodes were used to culture this mite. These species were Diplogaster sp., Panagrolaimus sp., Rhabditella leptura, Rhabditis elongata, Rhabditis teres, Phangrellus redivivus, Aphelenchus avenae, and Aphelenchoides composticora (Filipponi and di Delupis 1963, Ito 1971, Rodriguez et al. 1962, Singh and Rodriguez 1966). Methods for mass producing nematodes have been studied by Singer and Krantz (1967) and Singh and Rodriguez (1966). House fly stadia beyond the 1st-instar larvae were hardly suitable because their integument was too hard for the

mite to pierce and the larval moved too quickly (Filipponi 1955, Kinn 1966, Peck 1969).

Among muscoid species, M. muscaedomesticae preferred Musca domestica and M. autumnalis eggs over those of Fannia canicularis. There was no significant preference between eggs of M. domestica and M. autumnalis (Singh et al. 1966). The feeding on \underline{F} . canicularis was also reported by Axtell (1963c), O'Donnell and Nelson (1967) and Steve (1959). M. muscaedomesticae also fed on the eggs of the stable fly, Stomoxys calcitrans, and Phormia regina. Stable fly eggs were not preferred by these mites and did not appear to supply satisfactory nutrients for reproduction (Kinn 1966). The other organisms that had been reported to be the prey of this mite were springtails, leavens (yeasts), oligochaetes (Filipponi and di Delupis 1963, Singer and Krantz 1967), the human botfly, Dermatobia hominis (Moya Borja 1981), eggs of the grasshopper, Aulocara elliotti (Thomas), and the beetle, Pachyta lamed (L.) (Butler 1964).

Among the prey, house fly eggs and nematodes provided higher nutrition than other species. Feeding on house fly eggs produced more progeny than 1st-instar larvae (Filipponi and di Delupis 1963).

The larval stage of this mite was generally reported not to feed. However, Rodriguez et al. (1962) reported it feeding on nematodes.

A low concentration of ammonia stimulated the mite to puncture and pierce more house fly eggs (Wallwork and Rodriguez 1963). However, starvation for over 24 hours would not stimulate more predation (Rodriguez and Wade 1961). Tarsi 1 were important in locating food, since they had an olfactory function (Farish and Axtell 1966). Advanced study by Jalil and Rodriguez (1970a) showed the apical setae of tarsi 1 were responsible for detecting the house fly eggs.

It was interesting to note that the macrochelid mite may be a phoretic host for their prey--nematodes (Poinar 1965) or acarid mites (Axtell 1969, Chant 1960).

Life History

The life cycle of M. muscaedomesticae consists of egg, larva, protonymph, deutonymph, and adult. The larval stage is 6-legged, while nymphal and adult stages are 8-legged. It reproduces arrhenotokously, with fertilized eggs developing into females and unfertilized eggs developing into males. The females conceal their eggs by laying them in cracks or holes in the substrates (Filipponi and di Delupis 1963, Pereira and de Castro 1945, Wade and Rodriguez 1961). At 70% R.H., the mite can survive temperatures ranging from 10 to 36 C. Reproduction was observed at 15.5 to 36 C. Optimum oviposition temperatures ranged from 28 to 36 C

(Filipponi and Petrelli 1967, Filopponi et al. 1971). Temperature influenced the development time, fecundity, and longevity. The latter 2 were also density dependent. Non-mated females tended to have greater fecundity than mated females. The progeny of mated females had significantly higher mortality than the progeny of non-mated females. In addition, the female was very resistant to starvation. Fed after 15 days' fasting, 4 females started a normal reproduction period fairly similar to that of 4 normally fed females.

Specific studies were carried out at 28, 30, 32, and 34 C, and 75% R.H. (Filipponi et al. 1971). Using frozen house fly eggs and nematodes, mean development time for the female in hours was 59.8 at 28 C, 42.3 at 30 C, 49.5 at 32 C, and 49.9 at 34 C. The male had a shorter development time: 59.0 at 28 C, 41.4 at 30 C, 46.3 at 32 C, and 43.9 at 34 C. Females also lived longer than males. In the 28 to 34 C range, temperature did not affect female longevity but did affect the fecundity. Both the number of female progeny per female and the number of female progeny per female per day were affected. Oviposition reached an early maximum and then rapidly fell off (it underwent a flattening and progressive lengthening at lower temperatures). The female laid 159.65, 132.05, 127.2, and 72.65 eggs in her lifetime or 19.81, 17,85, 17.30, and 10.71 eggs per day at 28, 30, 32, and 34 C, respectively. The

parameters for the capacity of a population to increase showed that the optimum temperature range for \underline{M} . <u>muscaedomesticae</u> was from 28 to 32 C. The capacity of increase reached its maximum at 30 C. This mite could multiply by a factor of 2.174, 2.474, and 2.307 at 28, 30, and 32 C, respectively.

The sex ratio in the population and the progeny oscillated. The sex ratio at birth (sex ratio in the progeny) was negatively correlated to the sex ratio in the population. However, the overall sex ratio showed no significant difference. Sex ratio (females/total adults) in the population was 0.758, while at birth it was 0.440.

Last, there were inter-strain differences in the fecundity and development time of this mite (Filipponi and Petrelli 1967, Filopponi et al. 1971).

Wade and Rodriguez (1961) studied the life history of M. muscaedomesticae at 80 F (=26.7 C) with 55-60% R.H. Their results were generally similar to Filipponi and Petrelli. The development times were 56.35 and 54.51 hours for females and males. The non-mated females laid significantly more eggs (91.7) than mated females (61.4). Eggs from non-mated females were smaller, 0.370 mm x 0.249 mm, than those from fertilized females, 0.374 mm x 0.256 mm, and had longer incubation periods and longer development times. The maximum recorded egg laying per day was 28 and 25 eggs

for non-mated and mated females respectively. Females lived longer than males. Sex ratio at birth was 0.433.

Annual Abundance

A yearlong or semi-yearlong survey on the population of this mite in poultry manure has been done by Peck and Anderson (1969) in northern California, Rodriguez et al. (1962, 1970) in Kentucky, and Axtell (1970a) in North Carolina. Their works showed similar seasonal succession for this mite. The mites were most abundant from June to October. This differed from research with cattle manure, in which M. muscaedomesticae peaked twice a year, once in the spring around March and April and once in the winter around November and January (Rodriguez et al. 1962, 1970).

Manure removal had a disastrous effect on predaceous mites and insects, including M.

muscaedomesticae, Fuscuropoda sp., O. leucostoma, and histerids. Their population declined to the lowest point after manure removal (Peck and Anderson 1970). Among the 6 poultry farms studied by Axtell (1970a), one farm had not removed any manure since early September of the year before. In this farm, M.

muscaedomesticae built up in early May and covered most of the fly season, in contrast to the other farms'

July-September peak.

The report of Willis and Axtell (1968) differed from reports of other workers, since they found this mite peaked 2 to 3 weeks after the complete removal of manure then declined to a lower level.

Mass Production

As Filipponi (1964) pointed out, the production of nematodes was quite easy, the culture methods for the house fly have been developed, and therefore, mass production of macrochelid mites should be highly feasible.

Rodriguez and Wade (1961) studied the nutrition of this mite at 80 F, 55-60% R.H. An organic substrate with pH near 7 was most desirable. Tests on different manure preparation and additives in addition to frozen house fly eggs showed ground steer manure was better than fresh manure. Adding either 5% fish meal or 10% soybean oil meal was helpful in producing more mites. The fly larval medium, either new or used (spent), was tested. Spent fly larval medium was better than new medium. In the substrate which consisted of spent fly larval medium + 5% fish meal + 10% soybean oil meal + frozen house fly eggs, the mites reproduced at a rate of 2.6 progeny per female mite per day. However, the best 2 results came from steer manure + 10% soybean oil meal + frozen house fly eggs at 80 F, 58% R.H., and the same substrate plus 5% liver powder at 80 F, 48% R.H.

They produced 4.15 and 4.35 progeny per female mite per day, respectively, during the 5 days' testing period.

Nematodes were discovered to be a natural food of this mite species. Three nematode species, Diplogaster sp., Panagrolaimus sp., Rhabditella leptura, were cultured from cow manure. R. leptura was the easiest to rear and the best food. Using spent fly larval medium and providing R. leptura daily for food, the reproduction rate was 12.11 progeny per female mite per day. Nematodes combined with frozen house fly eggs gave 23.11 progeny per female mite per day. On fermented fly larval medium, nematodes alone gave as many progeny as both house fly eggs and nematodes. Yet pre-seeding the nematode resulted in fewer mite progeny (Rodriquez et al. 1962). Because of the value of nematode in rearing this mite, its mass production was studied. Singh and Rodriguez (1966) mass reared R. leptura with CSMA fly larval medium. Singer and Krantz (1967) cultured oligocheates and Rhabditis nematodes with crushed oats.

Ito (1973) studied the effects of nematode feeding on the reproductive rate of this mite at 27 C and 75-85% R.H. in a period of four days. He found that the reproductive rate of this mite was largely affected by the number of nematodes, Rhabditis elongata (Schneider), and/or house fly eggs. He compared the food value of house fly eggs, nematodes, and both. When adding house fly eggs once only at the beginning of the experiment, mites fed on nematodes alone gave the

highest reproductive rate, 15.2 progeny per female mite per day. Mites fed on nematodes plus a daily supply of house fly eggs had a reproductive rate slightly higher than when fed on nematodes alone. Generally, nematodes possessed a higher food value than house fly eggs. In another study, Ito (1977b) reared this mite with nematodes only. Nematodes, Rhabditis sp., were supplied every five days (treatment 1) or only at the beginning of the experiment (treatment 2). Number of progeny per females at the 5th and 10th day showed no difference between these two treatments. Progeny produced at the 5th and 10th day were 109.6 and 163.6 for treatment 1 and 109.6 and 164.4 for treatment 2. In the 15th and 20th day, treatment 1 had more progeny. The proportion of female adults in the population increased with time. At the 15th day 60.1% and 77.5%, and at the 20th day 89.3% and 98.8% of the population of treatment 1 and 2 were females, respectively.

Another factor affecting reproduction rate was ammonia. After reaching a certain concentration, ammonia could be harmful to the reproduction of this mite (Wallwork and Rodriguez 1963).

Predation/Control on House Fly

The predation rates of \underline{M} . $\underline{muscaedomesticae}$ on fly eggs and larvae will vary according to the technique and especially the substrates used.

Laboratory tests

In closed vials with no food other than fly eggs, Axtell (1961a) determined the predation rate of M.

<u>muscaedomesticae</u> to be 2.7 house fly eggs and 1st-instar larvae per female mite per day over a 3-day period. Following the same technique for 7 days, 0'Donnell and Axtell (1965) observed a consumption of about 3.5 eggs and 1st-instar larvae per female mite per day. Rodriguez and Wade (1961) obtained a higher rate on filter paper (19.8 eggs/female mite/day). But the test was conducted for only 24 hours and used mites which were previously starved for 36 hours.

Filipponi (1955) collected female adults of this mite from house flies. Inoculating with eggs or lst-instar larvae of the house fly at different prey/mite ratios, the number of matured larvae or adults were counted later and compared against a control. At the ratio of 5/1, 10/1, and 15/1, the mite killed 86.6%, 76.5%, and 73.1% fly eggs, respectively. At the ratio of 5/1 and 15/1 the mite killed 52% and 56.7% lst-instar fly larvae. The contact time was 2 hours for a ratio of 5/1, and 9 hours for the ratio of 10/1. The number of prey and mites were 50 prey/10 mites, 100 prey/10 mites and 300 prey/20 mites. Except for the test with 100 fly eggs/10 mites, the mites were starved after capture for 3 days for the test of 50 fly eggs/10 mites and 2 days for the remaining tests.

Rodriguez and Wade (1961) starved female adult mites and then tested their predation on house fly eggs on different substrates. These substrates were filter paper, steer manure plus 10% soybean oil meal, new fly larval medium, and spent fly larval medium. Five mites were added to the substrate with 250 fly eggs in each treatment. On the varied substrates 39.5%, 16.9%, 44.3%, and 34.6% of the fly eggs, respectively, were killed by the predation of mites which had been starved for 26 hours. For those mites which had been starved for 60 hours the results were 22.1%, 16.7%, 29.6%, and 68.2%, respectively. Except in the case of spent fly larval medium, a longer starvation period did not provoke more predation. Kinn (1966) found that M. muscaedomesticae could destroy about 3 to 4 S. calcitrans eggs, 6 to 10 P. regina eggs, or up to 7 house fly eggs in a day. King (1964) added 45 adult female mites to 250 house fly eggs. Based on larval survival, 80% of the eggs were killed. In the second test the mortality increased to 85%. Peck (1969) fed 10 adult female mites with 200 house fly eggs on the CSMA fly larval medium. There were 64.6 fly pupae recovered after 9 days with consumption of 61.4% of the fly eggs by the mites.

In a comparison test with Fuscuropoda vegetans, $\underline{\text{M}}$. <u>muscaedomesticae</u> produced higher house fly mortality. Using a ratio of 100 fly eggs to 20 mites (adult female

M. muscaedomesticae or unsexed adults or deutonymphs of F. vegetans) and then counting the 3rd-instar fly larvae after 72 hours, M. muscaedomesticae, F. vegetans adults, and F. vegetans deutonymphs resulted in 95.4%, 24.6%, and 5.6% mortality, respectively, on the production of house flies (O'Donnell and Axtell 1965). Two hundred and fifty house fly eggs and a varied number and species of 36-hour-starved mites were added to CSMA fly larval medium. The 3rd-instar fly larvae were counted later to determine fly mortality. Ten adult female M. muscaedomesticae plus 20 unsexed adult F. vegetans produced 55.7% mortality. Statistically, this was not significantly different from the mortality caused by 10 adult female M. muscaedomesticae (48.4%), but signifacantly different from the mortality caused by 20 adult F. vegetans (14.8%). The predation on the eggs of F. canicularis by M. muscaedomesticae and F. vegetans was also studied by O'Donnell and Nelson (1967). Each adult female $\underline{\text{M}}$. $\underline{\text{muscaedomesticae}}$ consumed 14.2 eggs on the average in the observed 5 days. Female and male adult F. vegetans ate 7.0 and 6.4 eggs in the observed period of 11 and 9 days, respectively.

The index of predation potential calculated by Peck (1969) was 95.8 for adult female $\underline{\text{M}}$. muscaedomesticae.

Field tests

Axtell (1963b, 1963c) demonstrated the reduction of house fly production in cow and chicken manure by the naturally occurring predaceous mites or introduced M. muscaedomesticae and G. confusa. Adding 20,000 house fly eggs to caged intact calf pen manure, the area with undisturbed mite population produced 61% to 67% fewer house flies than the area with the mite population destroyed by dicofol. When tested on outdoor dairy cattle manure piles with the addition of 50,000 house fly eggs, 31% to 45% fewer house flies were produced by the area with undisturbed mite populations. Tested with indoor caged fresh dairy cattle manure to which 20,000 house fly eggs were added, 94% fewer flies were produced by the area that had 200 M. muscaedomesticae and 200 G. confusa added. Similar results were obtained three weeks later when 20,000 and 60,000 eggs and fresh manure were added to the same manure piles. In caged-layer droppings, a natural population of M. muscaedomesticae was allowed to build up for 3 weeks. Four thousand house fly eggs were then added to the droppings in pans under cages. There were 80% and 83% fewer house flies and little house flies (F. canicularis) produced. The addition of 50 adult female G. confusa could cause 51% destruction of 40,000 house fly eggs.

Introducing \underline{M} . \underline{M} . \underline{M} muscaedomesticae and house fly eggs approximately in the ratio of 1/5, Singh et al. (1966)

observed an 85.5% control on the house fly in cow manure, 91.9% control on the little house fly in poultry manure, and 23.4% control on M. autumnalis in cow manure. Rodriguez et al. (1970) studied the control of the house fly (1000 eggs) by the addition of 200 mites (either M. muscaedomesticae or F. vegetans or both species) and the naturally occurring mites in fly-isolated containers that were set in a poultry house. M. muscaedomesticae, F. vegetans individually and in combination gave 99%, 87% and 92% control of the house fly. Ito (1973) offered both house fly eggs and nematodes to M. muscaedomesticae in the laboratory mite medium. The predation rate of these mites on house fly eggs was influenced by the presence of nematodes: the more nematodes, the lower the predation rate. But tests on fresh pig manure showed no difference in the predation on house fly eggs whether the nematodes were present or absent (Ito 1977a).

The phoretic behavior of this mite on the house fly can shorten the life span of the adult house fly. This may also result in reduced egg laying and an additional control to the predation on eggs and lst-instar larvae (Kinn 1966).

Integrated Control Program

The integrated control of the house fly by $\underline{\mathsf{M}}$. $\underline{\mathsf{muscaedomesticae}}$ and other agents has been studied by Axtell (1966, 1968, 1970a, 1970b), Peck and Anderson (1970), Rodriguez et al. (1970), and Wicht and Rodriguez (1970); and it has also been discussed by Anderson (1983), Krantz (1983), and Singh and Rodriguez (1969).

Axtell (1966) studied the relative toxicity of 17 insecticides to the 3rd-instar larvae of the house fly and adult female mites. The mites and fly larvae were exposed to insecticides incorporated in the CSMA fly larval medium. Propoxur, dichlorvos, fenthion, malathion, crotoxyphos, and naled were more toxic to the mites than to the fly larvae. Dimetilan and chlordane were more toxic to the mites at the LC95 level and more toxic to the fly larvae at the LC50 level. Diazinon was about equally toxic to the mite and the fly larvae. DDT, trichlorfon, and ronnel were slightly more toxic to the fly larvae than to the mites. Chlordecone, dimethoate, lindane, GC 9879,* and coumaphos were more toxic to the fly larvae than to the mites.

Chlordecone and dimethoate had the greatest selectivity. Continuing the work, 12 insecticides were selected and applied to poultry manure. The 3rd-instar larvae of the house fly and adults of this mite were determined at intervals before and after insecticide

^{*} experimental insecticide

application. Selective toxicity in favor of the mites was not found. Insecticides which did not destroy the mite population were not effective in controlling the fly larvae. Those which gave control to fly larvae were deleterious to the mite population. The mite population increased very slowly after decimation by the insecticide application while the fly larvae increased rapidly. It is concluded that non-selective insecticides should not be applied to the manure. Selective application to control adult house fly should be intensively studied (Axtell 1968), Axtell (1970a) compared the different fly control programs in caged layer farms. Populations of house flies, little house flies, black garbage flies, and predaceous manure-inhabiting mites (Macrochelidae, Parasitidae and Uropodidae) were determined weekly from March/April till September. Excellent fly control resulted from a program based on early season manure removal and the control of adult house fly by insecticide-bait stations and 5 or 6 selective applications of insecticide to the inside upper parts of the poultry house and the interior and exterior surfaces of the attached feedand egg-storage buildings. In another study, Axtell (1970b) compared the effect of weekly larviciding of the manure with RaVap or Zytron to the integrated selective adulticiding of the inside upper part of the poultry house by RaVap in caged-layer farms. Both

methods gave the same satisfactory control on the house fly and little house fly. But the larviciding of manure would require 5 times as much insecticide and 2.5 times as many man-hours per season as would the adulticiding program. Larviciding at biweekly intervals did not give satisfactory fly control.

Rodriguez et al. (1970) studied the relative toxicity of 14 insecticides to the newly hatched house fly maggots and adult female mite. Ronnel, diazinon, malathion, dimethoate, Bayer 38156* and CIBA-9491* were more toxic to the fly larvae than mites. In the follow-up study, diazinon, ronnel, CIBA-9491, and malathion at the LC50 range and a mite/house fly egg ratio of 1/20 were tested on a substrate of 5 gm CSMA fly larval medium plus 20 gm poultry manure. The experimental results indicated that mites plus insecticide were most effective; mites alone were less effective; and insecticide alone was the least effective. Another approach was the use of baits (trichlorfon and bomy1 baits), which showed promising results for integrated fly control.

Wicht and Rodriguez (1970) studied the integrated control of the house fly and the little house fly using predaceous mites, selective insecticides, and microbial agents. As tested in the laboratory, fenthion, abate,

^{*} experimental pesticides

coumaphos, ronnel, diazinon, Kepone, and C-9491 were relatively more toxic to house fly larvae than to M. muscaedomesticae. Bacillus thuringiensis and the parasitic nematode, Neoplectana carpocapsae, were harmless to the mite. Tested in the caged-layer house, all 7 insecticides decreased the mite populations of M. muscaedomesticae and F. vegetans. B. thuringiensis was most effective against fly larvae. Liquid poisoned baits were also studied. They provided effective

Peck and Anderson (1970) studied the influence of the poultry manure removal schedule on various dipteran larvae and selected arthropod predators. The population of Fannia spp. increased after the weekly, biweekly, or monthly removal of manure faster than O. leucostoma, M. muscaedomesticae, Fuscuropoda sp., and histerid beetles. The 3rd-instar larvae of the house fly, the false stable fly, and calliphorids were most abundant in 1-week-old manure; those of F. canicularis, F. femoralis, and the black garbage fly dominated the 2-to 3-week-old manure. Unremoved manure had the least number of dipterous larvae except for the stable fly.

Axtell (1981) suggested that filth flies should be controlled through a fly-management program. The program should include 1. manure management: keep manure dry, remove manure after a long interval, and leave a thick base of old manure to perpetuate natural

enemies; 2. selective use of insecticides: use selective insecticides and apply in a selective way or with a selective formula; 3. augmentation of natural enemy populations: will need studies to determine the manner and the time. This management program should be part of the IPM of all poultry pests.

Anderson (1983) reviewed the works on biocontrol of dung-breeding pests by mites, mainly $\underline{\mathsf{M}}$. $\underline{\mathsf{muscaedomesticae}}$ and $\underline{\mathsf{F}}$. $\underline{\mathsf{vegetans}}$, and considered the selection of insecticide-resistant mites that are predaceous on filth flies. Krantz (1983) reviewed the works on dung-associated macrochelid mites. He pointed out that in order to favor the macrochelid mites to control domestic dung flies, the following considerations may be necessary:

1. Sanitation practices tend to eliminate the mites.

Manure should not be removed completely. 2. An IPM
program should be beneficial. However, better
understanding of the complex interactions between and
among the biotic and abiotic components of the dung
system is required for IPM. 3. High research priority
should be given to the selection of pesticide-resistant
strains of coprophilous predaceous mites. 4. An
artifical mass-rearing method of macrochelid mites is
needed. 5. The potential of using fimicolous
(dung-inhabiting) mites not normally found in domestic
dung should be studied.

CHAPTER 3

POPULATION FLUCTUATION OF SELECTED ARTHROPODS IN POULTRY MANURE

Materials and Methods

The study on seasonal abundance of Macrocheles muscaedomesticae was conducted at a commercial caged layer farm in Bradford County, Florida. The farm consisted of 5 open-sided houses oriented east-west. Each house had 4 rows of wire cages suspended 1 m above the ground. Two of the rows were joined back to back along the central axis of each house, and 2 additional rows were similarly located, one along each of the sides. The poultry droppings that accumulated beneath the 2 central rows of cages were separated from the droppings beneath the 2 outer rows of cages by a concrete aisle. Each row had 350 cages with each cage containing 4 layers, giving a total of 5600 layers in each house, or 28,000 layers on the farm. The droppings beneath the southern central row of the second house on the south were selected for sampling (Figure 3-1). On each end, the droppings beneath 25 cages were ignored because of the recurrent water accumulation at one end. The remaining 300 cages were divided into 20 sections, each containing 15 cages. Twenty manure



Figure 3-1. The sampling poultry house.

samples, one from each section, were taken weekly from 23 September through 28 October, 1983, and then biweekly through September 7, 1984. Cages in each section were numbered 1 to 15 beginning at the eastern end. A number between 1 and 15 was chosen from a random-digit table before sampling, and the manure beneath the cage of this number was sampled. For example, if the random number selected was 8, then a sample was taken under cage 8 in each section of 15, for a total of 20 samples.

Each sample consisted of approximately 300 \mbox{cm}^3 of manure, collected with a trowel and packed in a sealed plastic container.

Although Willis and Axtell (1968) found M.

muscaedomesticae mites were distributed primarily on
the outermost layer and near the peak of the manure, in
my preliminary sampling, they were not found in the
dried droppings. Therefore, mainly fresh droppings were
collected, or, when the manure was too wet, then nearby
manure which was slightly drier was collected. The
macrochelid mites appeared to distribute themselves
around the inner boundary of the mixture of chicken
feed and soil beneath the cages when the droppings
beneath a systematically selected cage were liquified
by the maggots of the soldier fly. Therefore, samples
were collected at the boundary between the watery and
dry areas. The surface layer within 2 cm was collected

if the entire area beneath that cage was watery. The

Samples were taken between the hours of 10 AM and 1 PM. The arthropods in the manure were separated by a Tullgren Berlese funnel into 99% isopropyl alcohol (Figure 3-2) within 3 hours after obtaining the sample to preclude the mortality of the M. muscaedomesticae in the tightly covered plastic container. Forty-watt light bulbs were used to drive manure-inhabiting arthropods down to the isopropyl alcohol. Three to four days, depending on the moisture level of the manure sample, were required to complete the separation. The number of adult female and male M. muscaedomesticae and the 3rd-instar house fly larvae were counted.

Since small particles of various materials often dropped down into the alcohol, the presence of these particles obstructed the counting of arthropods. It was then necessary to drain the alcohol and added a 10% saturated sugar solution to the vial. Arthropods floated by the sugar solution were poured over the C-299 maiden chiffon and counted.

To understand the relationship between the manure moisture level and the distribution of $\underline{\mathbf{M}}$. $\underline{\mathbf{muscaedomesticae}}, \text{ the manure moisture level was}$ $\underline{\mathbf{measured}} \text{ from the beginning of the study till July 1},$ 1984. Falcon 1007 disposable plain petri-dishes were $\underline{\mathbf{used}} \text{ for measuring the moisture level in the chicken}$



Figure 3-2. Tullgren Berlesi funnels for separating arthropods.

manure. One petri-dish of manure was taken from each manure sample, weighed, and oven-dried at 50 C for 3 days, and then weighed again. The moisture level was calculated as follows:

wet weight - dry weight
moisture level = ------ x 100%
wet weight

From October 21, 1983, the temperature of air and manure sample were measured before sampling. Using a mercury thermometer, two measurements were taken, one on each end of the poultry house. The manure temperatures were read 5 minutes after the thermometer was inserted into the manure.

Numerous insects and mites other than \underline{M} . $\underline{Muscaedomesticae}$ were found in chicken manure. They may be the competitors, natural enemies, or prey of \underline{M} . $\underline{Muscaedomesticae}$. No matter what their role, they might influence the population abundance of \underline{M} . $\underline{Muscaedomesticae}$. In order to gain information on their importance, the abundance of the following groups of insects and mites in the samples were described from February 24, 1984, until termination of sampling:

Insects:

- B: beetles and their larvae, including Histeridae, Staphylinidae, and Tenebrionidae.
- F: Larvae of Fannia spp.

M: young muscoid maggots, including those of the house fly and those similar to house fly.

Mites:

- A: Acaridae.
- S: Sejoidea.
- U: Uropodidae.
- O: Other mites.

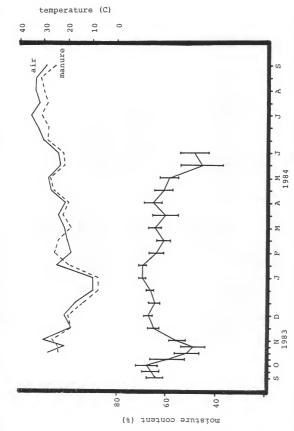
Three categories were used for describing the abundance of them:

- 5. Abundant
- 3. Moderate
- 1. None to poor

The absolute abundance of each category was varied in different groups. For example, the actual number of "abundant" B was far less than "moderate abundant" or even "poor" mites of any group. In other words, this observation described the relative population fluctuation of only each group against itself.

Results and Discussion

Figure 3-3 shows the temperature of air and manure and the mean and 95% confidence intervals of the percent moisture level of manure samples. The temperature was generally between 20 to 30 C. However, there was a time period, December and January, when it



Fluctuations of temperature and moisture content in caged-hen manure. Figure 3-3.

dropped below 20 C. Especially on December 30, 1983. and January 13, 1984, when the temperature was 8 and 10 C in the air and manure respectively. The lowest temperature recorded around that time was reported to be -8.3 C by the U.S. weather service. The manure temperature exceeded 30 C in the months of June, July, and August. It reached a maximum of 35 C on July 13. Previous sampling had not collected M. muscaedomesticae from watery manure and rarely collected it from rotovated or disced dry manure. Peck and Anderson (1969) collected this mite species most frequently from manure that had a moisture level of about 63%. The moisture level of manure was, then, an important factor that affected the distribution of this mite species and, therefore, was measured. The moisture level of manure was in the 60% range most of the time. It dropped to below 50% twice, first at the end of October 1983 and second in May and June 1984. The moisture level of manure shows high uniformity by its narrow 95% confidence interval.

The manure was removed around October 27, 1983. The insecticide schedule for the control of the house fly larvae in the manure was Ectiban 5.7% E.C. on November 1, 1983, and April 14, 1984; dimethoate 23.4% E.C. on May 27, 1984. Then Ectiban 5.7% E.C. was spot-sprayed at the place where the house fly larvae were abundant.

Figures 3-4 and 3-5 show the mean number and 95% confidence interval of the 3rd-instar house fly larvae and adult \underline{M} . \underline{M} muscaedomesticae, respectively, in 20 chicken manure samples. Based on the observations on feeding amount and the sex ratio in the population, the researcher considers the abundance of the adult female mites as the most meaningful indicator for the seasonal abundance of \underline{M} . \underline{M} muscaedomesticae.

Both populations showed a small peak in September and October 1983. At the beginning of October, the farmer was providing the hens with water only, forcing them to molt. The subsquent decline of both populations in October may be related to the molt of the hens.

The manure removal around October 27, 1983, provided a disastrous effect upon both the mite and house fly larvae populations. The population of the 3rd-instar house fly larvae dropped to zero on November 28, 1983, and jumped back to peak 7 days later. This was also observed by Peck and Anderson (1970). Seven days was the approximate development time for the house fly from egg to 3rd-instar larvae. Therefore, this peak represents an outbreak of the first generation of the house fly larvae after the manure removal. As there was no reason for an outbreak of adult flies around the manure removal, it could only be explained on the basis of lack of natural control agents. However, the population of the M. muscaedomesticae did not recover

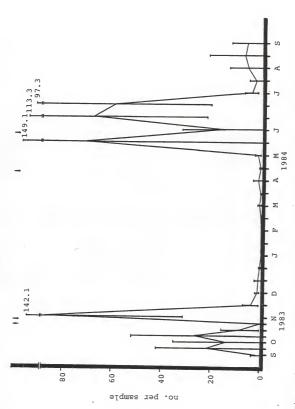


Figure 3-4. Population frequencies of the 3rd-instar house fly larvae in caged-hen manure.

insecticide applications
 manure removal

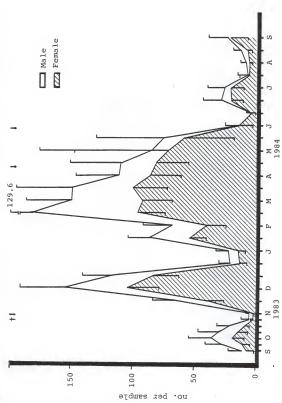


Figure 3-5. Population frequencies of M. muscaedomesticae in caged-hen manure. + insecticide applications + manure removal

as soon as the house fly larvae. The mites peaked 4 weeks later than the house fly larvae. The time delay was the result of the combined effect of the time needed for the mature larvae to develop into adult house flies and to oviposit, and the usual delayed density dependence between predator and prey. The time lag could be shorter than 4 weeks because samples were taken biweekly. The spray of Ectiban 5.7% E.C. on November 1, 1983, may also have affected the delay of the buildup of the mite population. It seemed to be more harmful to the mite population than to the fly larvae.

The mean number of fly larvae per sample dropped to below 2.0 after the buildup of the mite population. The fly larval population remained low until May 1984. In the meantime, the population of M. muscaedomesticae reached a plateau, except for the single drop that was caused by the severe cold from December 30, 1983, to January 13, 1984.

The cold weather seemed to be more hazardous to the fly population than to the mite population. No 3rd-instar fly larvae were collected in January and Febuary 1984. Yet, M. muscaedomesticae was not eliminated by the cold weather. The mites averaged 12.9 and 14.4 per sample on December 30 and January 13, respectively, and its population subsequently increased. Studies by Butler (1964) indicated that this

mite insulated itself by manure layering and consequently survived the severe temperatures in the northwest.

The fly larval population peaked again during May and June of 1984, dropped to a low level in July, and remained low until the end of the yearlong survey. The application of Ectiban on April 14, 1984, seemed to have no effect on the population of $\underline{\mathbf{M}}$. $\underline{\mathbf{Muscaedomesticae}}. \ \, \mathbf{However}, \ \, \mathbf{the} \ \, \mathbf{application} \ \, \mathbf{of} \ \, \mathbf{dimethoate} \ \, \mathbf{on} \ \, \mathbf{May} \ \, \mathbf{27}, \ \, \mathbf{1984}, \ \, \mathbf{reduced} \ \, \mathbf{both} \ \, \mathbf{the} \ \, \mathbf{mite} \ \, \mathbf{and} \ \, \mathbf{the} \ \, \mathbf{fly} \ \, \mathbf{larval} \ \, \mathbf{populations}. \ \, \mathbf{The} \ \, \mathbf{fly} \ \, \mathbf{larval} \ \, \mathbf{recovered} \ \, \mathbf{rapidly}, \ \, \mathbf{but} \ \, \mathbf{the} \ \, \mathbf{mites} \ \, \mathbf{remained} \ \, \mathbf{at} \ \, \mathbf{lower} \ \, \mathbf{levels} \ \, \mathbf{until} \ \, \mathbf{September}.$

Generally, the population of M. muscaedomesticae was negatively correlated to the population of 3rd-instar house fly larvae, except in September and October in 1983, and from July to September in 1984. A comparison of the manure moisture content (Figure 3-3) showed that the population of this mite dropped when the manure moisture content was lower than 50%. This happened twice, first in October 1983 and then in May 1984. On May 18, 9 manure samples of the 20 samples taken had a moisture content that exceeded 55%. If these 9 samples are not considered in total analysis, the mean number of mites in the remaining 11 samples is 14.2 rather than 58.1. The distribution of this mite in

different moisture levels of caged-layer manure is analyzed in Table 3-1.

The distribution of the number of mites per sample, the total number of mites, and the number of manure samples that contained these mites all showed the same result: these mites preferred the manure that had a moisture content of 65.0-69.9%, followed by a moisture level of 60.0-64.9%, and then (approximately equally) the moisture level of 70.0-74.9% and 55.0-59.9%. Beyond these ranges, the dry manure appears to be more tolerable than the wet manure. Therefore, the moisture content of the caged-layer manure seems to be the key factor in limiting the distribution of M. muscaedomesticae. This mite prefers caged-layer manure with a moisture content of 55.0-74.9%, with a 60.0-69.9% moisture content being the optimum. This range covers the ideal moisture level (65-70%) for this mite which was reported by Rodriguez et al. (1962).

The preference for manure moisture level by \underline{M} . <u>muscaedomesticae</u> is more apparent from the distribution of the total numbers of this mite than from the distribution of the number of manure samples. These are compared in Figure 3-6. At a moisture level of 60.0-69.9%, 210 (51.3%) manure samples contained 20,230 (65.3) mites, or 88.3% of the mites were collected in the manure with a moisture level of 55.0-74.9%.

Table 3-1. Distribution of $\underline{\text{M}}\text{.}$ $\underline{\text{muscaedomesticae}}$ in different moisture levels of caged-hen manure.

Moisture	Manure	samples		Mites	
level(%)	No.	8	Total	no. %	No./sample
0-39.9	10	2.4	138	0.4	13.8
40-44.9	13	3.2	269	0.9	20.7
45-49.9	22	5.4	899	2.9	40.9
50-54.9	35	8.6	1238	4.0	35.4
55-59.9	51	12.5	3127	10.1	61.3
60-64.9	95	23.2	8453	27.3	89.0
65-69.9	115	28.1	11777	38.0	102.4
70-74.9	60	14.7	3979	12.9	66.3
75-79.9	8	2.0	1081	3.5	135.1(28.0)*

^{*} If the sample with 885 mites is not counted.

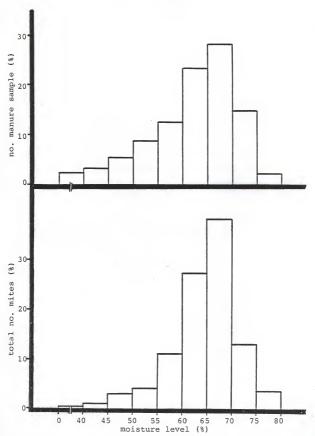


Figure 3-6. Distribution of $\underline{\text{M.}}$ $\underline{\text{muscaedomesticae}}$ in different moisture level of caged-hen manure.

M. muscaedomesticae was collected from manure with a moisture content as high as 77.6% and as low as 14.7%, and the most numerous mites collected in one sample were from the manure which had the highest moisture content, 77.6%. The manure mass with a moisture content lower than 45% rarely contained this mite. In general, caged-layer manure with a moisture content over 73% is black in color and anaerobic, unsuitable for the mites. This is valid if one does not count the one manure sample which had an extraordinarily high number of this mite, 885. Two factors were considered by the researcher to be the reason for the presence of this mite in the very dry and anaerobic manure. First, house fly larvae and eggs do occur in both the dry and anaerobic area as indicated by Figure 3-7 and Table 3-2. There is usually a drier surface layer on the anaerobic manure. Therefore, the presence of the food may have an effect beyond the moisture content of manure on the distribution of this mite. Miller et al. (1974) reported that at 27 C, the poultry manure with a moisture content of 60-75% was suitable for house fly larval development. Manure becomes anaerobic and unsuitable for house fly larval development when its moisture content exceeds 80%. This is slightly different from the observation of this researcher. An analysis of the distribution of the 3rd-instar house fly larvae in caged-layer manure at

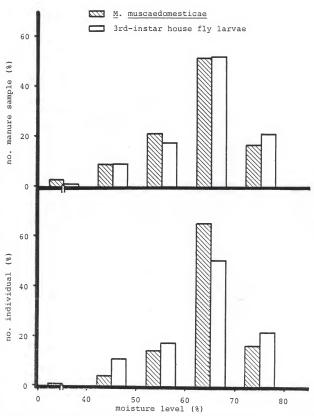


Figure 3-7. Distribution of the \underline{M} . $\underline{\underline{M}}$ $\underline{\underline{M}$ $\underline{\underline{M}}$ $\underline{\underline{M}$ $\underline{\underline{M}}$ $\underline{\underline{M}$

Table 3-2. Distribution of 3rd-instar house fly larvae in different moisture levels of caged-hen manure.

Moisture	Manure	samples		L3*	
level(%)	No.	8	Total r	10. %	No./sample**
0-39.9	1	0.7	2	0.04	
40-44.9	1	0.7	73	1.5	
45-49.9	11	8.0	449	9.1	43.5
50-54.9	10	7.3	289	5.8	
55-59.9	14	10.2	567	11.5	35.7
60-64.9	33	24.1	1187	24.0	
65-69.9	38	27.7	1304	26.4	35.1
70-74.9	24	17.5	242	4.9	
75-79.9	5	3.6	833	16.8	37.1

^{* 3}rd-instar larvae.

^{** 10%} moisture level groups.

different moisture levels is shown in Table 3-2. The distribution of these larvae was similar to \underline{M} . muscaedoesticae: high numbers in manure with a moisture level of 60.0-69.9%, lower populations in other moisture levels.

The percentage distribution of these larvae in different manure moisture levels (in classes of 10%) is compared to \underline{M} . Muscaedomesticae in Figure 3-7.

There was an approximately equal probability of finding M. muscaedomesticae or 3rd-instar house fly larvae in any of these manure moisture levels. But M. muscaedomesticae was more numerous than house fly larvae in manure with a moisture level of 60.0-69.9% and less in other manure moisture levels. The mean number of 3rd-instar house fly larvae per sample in each manure moisture level was very close. This may be because either the house fly distributed evenly in manures of different moisture levels or the varied density of M. muscaedomesticae in manures of different moisture levels allowed approximately equal density of house fly larvae. Nevertheless, the food did not seem to be a key factor in limiting the distribution of the mite, although in some cases it did surpass the effect of the manure moisture level on the distribution of the mite.

Another food factor that may be more important than the house fly eggs and lst-instar larvae is the

rhabditid nematodes. Nymphs of \underline{M} . $\underline{muscaedomesticae}$ selectively prefer the nematode for food. Nematodes are commonly found in poultry manure. Their effect on the distribution of this macrochelid mite is not known.

The fluctuations of 7 arthropod groups from
February 24 to September 7, 1984, are shown in Figure
3-8 (3 insect groups) and Figure 3-9 (4 mite groups).
Fluctuations of any 2 groups are not comparable because
their abundance was determined on different standards.
This study is preliminary and rough. More studies are
needed to substantiate it.

Beetles and larvae of Histeridae, Staphylinidae, and Tenebrionidae are common predators found in poultry manure (Bill 1973, Legner 1971, Peck 1969, Peck and Anderson 1969, Pfeiffer and Axtell 1980). Larvae of Tenebrionidae (the mealworm) had been reported to give good control of the house fly in a poultry farm (Hartman 1970). Their fluctuation (Figure 3-8, B) peaked in May and June, the same time period of the 2nd peak of the 3rd-instar house fly larvae. However, they are probably not important in the control of the house fly in Florida because of the low density of their population in comparison with the number of 3rd-instar house fly larvae present. Though M. muscaedomesticae was reported to feed on the eggs of Fannia canicularis (Axtell 1963c, O'Donnell and Nelson 1967, Singh et al. 1966 and Steve 1959) and can control this fly (Singh et

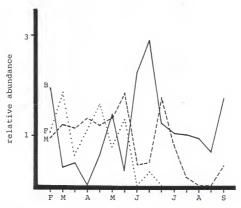


Figure 3-8. Frequency distribution of selected insect groups in caged-hen manure.

B: Beetles and their larvae,

F: Fannia spp., larvae,

M: Young muscoid maggots.

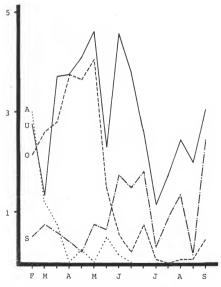


Figure 3-9. Frequency distribution of selected mite groups in caged-hen manure.

A: Acaridae

S: Sejoidea

U: Uropodidae

O: Other mites.

al. 1966), the fluctuation of the larval population of Fannia spp. (Figure 3-8, F) was not related to $\underline{\mathsf{M}}$. $\underline{\mathsf{muscaedomesticae}}$. The study of Axtell (1970a) also showed the same result. The peaking of young muscoid maggots (Figure 3-8, M) seemed to be synchronized with the 3rd-instar house fly. The population of young maggots outnumbered the mature maggots before May. The lst-instar larvae among them served as the food for predaceous mites.

The population of acarid mites dropped in March and remained low until the end of this study (Figure 3-9, A). They were abundant in fresh manure and generally were saprophagous. They might be a food source for M. muscaedomesticae, if M. muscaedomesticae can feed on them. The population of sejoid mites fluctuated in a similar manner to the beetles (Figure 3-9, S). They were numberous in caged-hen manure. The food habits of sejoid mites are unknown. The uropodid mites were the most abundant arthropods in the manure. Uropodids fluctuated in a pattern similar to beetles (Figure 3-9, U). They peaked from March to June with a drop on May 18, declined at the end of June, and then increased in July until the end of the study. Their decline at the end of June was probably caused by the application of insecticides. Therefore, they peaked and filled in the gap that was left by $\underline{\mathsf{M}}$. $\underline{\mathsf{muscaedomesticae}}$ during the summer months. Although a predaceous

uropodid mite, <u>Fuscuropoda vegetans</u>, was reported to be common in poultry manure (Jalil and Rodriguez 1970b, O'Donnell and Axtell 1965), this study showed that the uropodid mites apparently could not suppress the population of the house fly. The majority of the group of "other mites" were gamasid mites. They peaked in March, April, and May, then dropped in mid-May to the end of the study. Their fluctuation is not related to the fluctuation of the house fly.

Table 3-3 shows the frequency of collection of the 7 arthropod groups in different caged-hen manure moisture levels. Fannia spp. and young muscoid maggots occurred in the same moisture level as the 3rd-instar house fly larvae. Acarid mites occurred in fresh manure. Mites in the group of "other mites" occurred mostly in the moisture level between 55.0-69.9%, similar to M. muscaedomesticae. They probably feed on fly eggs, too. Sejoid mites occurred most frequently in the moisture level of 55.0-59.9%. The distribution of two known predaceous groups, uropodids and beetles and their larvae, was in a manure at moisture levels approximately 5% drier than the distribution of M. muscaedomesticae. This substantiates the observation of Peck and Anderson (1969). They found the poultry manures with the moisture content between 50% to 70%, from drier to wetter, were the foci of Histeridae, Staphylinidae, Fuscuropoda sp., and M. muscaedomesticae.

Table 3-3. Frequency of collection (number of manure samples) of selected arthropod groups in different moisture levels of caged-hen manure.

 Moisture level (%)	A	s	U	0	В	F	М	
0-39.9	1	2	7	3	4	0	1	
40-44.9	2	4	9	7	3	0	0	
45-49.9	0	4	9	5	7	4	5	
50-54.9	0	4	11	10	5	2	7	
55-59.9	1	8	30	24	14	0	16	
60-64.9	7	5	26	28	14	8	27	
65-69.9	8	3	22	23	9	14	26	
70-74.9	5	0	6	8	0	8	9	
 75-79.9	0	0	1	2	0	4	3	

A: Acaridae

B: Beetles and larvae of Histeridae, Staphylinidae and Tenebrionidae

F: Fannia spp.

M: Young muscoid maggots

O: Other mites, mostly gamasids

S: Sejoidea

U: Uropodidae

The results of this study indicate the negative density dependence between the population of $\underline{\mathbf{M}}$. $\underline{\mathbf{muscaedomesticae}}$ and the 3rd-instar house fly larvae and that the house fly larvae and the mites have the same preference in manure moisture level. The extremely low house fly larval population during the winter and spring of 1983-1984 as well as the unimportance of other predators, all strongly suggest that the $\underline{\mathbf{M}}$. $\underline{\mathbf{muscaedomesticae}}$ is the key predator in the control of house fly in poultry farms in Florida. However, this study was done on one poultry farm in part of one flock season. It was only one observation. More studies are needed to confirm the findings of this study.

Besides, these mites peaked 2-3 weeks after manure removal in the study of Willis and Axtell (1968) and 3 months after in the study of Axtell (1970a), and these mites were most abundant in summer in the studies of other researchers (Axtell 1970; Peck and Anderson 1969; Rodriguez et al. 1962, 1970). During this yearlong study, house fly larval populations escaped the control of these mites twice, once after the manure removal and once in summer. These show that the season of year and geographic location had an effect on the performance of this mite. In addition, studies should be done on the reasons for the failure of these mites to suppress house flies in the summer and on the effect of manure removal in different seasons and at different flock

ages. Other research should also include more than one poultry farm and possibly, the life span of the flock of laying hens.

CHAPTER 4

MASS PRODUCTION OF MACROCHELES MUSCAEDOMESTICAE AND STUDIES OF MODELING

Materials and Methods

Adult female mites, Macrocheles muscaedomesticae, were collected in 1982 from the caged-hen manure in the poultry houses of the Department of Poultry Science, IFAS, University of Florida, Gainesville, Florida. Six laboratory colonies were established, each from a single adult female. Male offspring of each colony were exchanged mutually among colonies for 2 months. Only one stock colony was maintained thereafter. Plastic containers, the Superseal brand 32-ounce oblong food saver and then the 80-ounce oblong food saver, were used to culture the mites. A hole of approximately 8.5 x 8.5 or 11.5 x 16.0 cm^2 was cut in the center of the lid for ventilation and sealed with C-299 maiden chiffon to prevent the dispersal of this mite and/or other arthropods. The plastic container was filled with spent house fly media (media remaining after house fly rearing) up to about 1 cm from the top margin of the wall. The constituents of this house fly media are given in Appendix 1. House fly eggs, previously chilled to prevent hatching, were supplied as the food for the

mites. Both the spent house fly media and the house fly eggs were obtained from the Insects Affecting Man and Animals Research Laboratory of USDA in Gainesville, Florida. The stock colony was reset weekly to maintain good quality and sanitation.

A nematode, Protorhabditis sp., was found to be abundant in the stock colony. This nematode can be mass produced very easily in the spent house fly media. Nematodes have been reported to be an excellent food for M. muscaedomesticae (Filipponi and di Delupis 1963; Ito 1971, 1977a; Rodriguez et al. 1962; Singh and Rodriguez 1966). Therefore, a colony of this nematode was also cultured in the laboratory. Widemouthed quart glass mason jars were used for the culture of the nematode. The glass jar was filled with spent house fly media and then 1-2 tablespoonfuls of media of the old nematode colony were added. The opening of the glass jar was covered by the same C-299 maiden chiffon and fastened with a rubber band. This type of closure provides ventilation and effectively isolates arthropods from invading the spent house fly media. At room temperature (21.1-26.7 C), the nematode reproduced in great numbers in 3 days. However, the abundance decreased after 5-6 days. Media from the nematode colonies (nematode media) of 3 to 5 days in age were supplied along with house fly eggs to the mite stock colony or to the experimental studies. The nematodes multiply on

the spent house fly media and consequently serve as the food for M. ${\tt muscaedomesticae.}$

The spent house fly media used in the studies in this dissertation were frozen before use for at least 3 days to kill unwanted organisms and then defrosted to room temperature right before use. The method of Morgan (1981a) was followed to measure the amount of house fly eggs in all experiments. The nematode media were measured with a 50 ml tri-pour disposable beaker made by Sherwood Medical Industries Inc. At the end of each test, the mites in each box were separated by a Tullgren Berlese funnel into 99.5% isopropyl alcohol and then counted. The separation of mites from 100 gm, 200 gm, and 400 gm or more of spent house fly media took 1, 2, and 3 or more days to complete. After the test of the effect of different foods on the reproduction of M. muscaedomesticae, the method of Ing (1978) (with a slight modification) was followed to estimate the number of M. muscaedomesticae. A 10% sugar solution was used (as described in Chapter 3) to separate this mite from the debris of spent house fly media that had fallen into the alcohol before the counting operation, since the debris would close the tip of the eye-dropper. All experiments in this chapter were conducted in an incubator at 30 ± 2 C, 60-98% R.H., and a photophase of 14:10 (L:D).

Mass Production of M. muscaedomesticae

The effect of different foods

The first factor to be tested was the food that should be supplied to the M. muscaedomesticae. The studies of Rodriguez and Wade (1961) showed that the addition of sovbean oil meal and fish meal could increase the number of progeny of this mite. The commercial layer feed and the Ferti-lome brand fish emulsion fertilizer 5-2-2 of Voluntary Purchasing Groups, Inc., Bonham, Texas, were used as the substitutes for soybean oil meal and fish meal. House fly eggs and nematodes were definitely to be tested because of their high nutritive value to this mite. A factorial comparison of the food value of the above materials was conducted. A 32-ounce box was filled with 100 gm spent house fly media. The amount of the testing materials added were house fly eggs, 1.0 ml; nematode media, 1/3 beaker; water, 30 ml (no water if fish emulsion fertilizer added); 0.1% solution of fish emulsion fertilizer, 30 ml; and commercial layer feed, 10 gm. Spent house fly media alone without the above additives was used as a check. 10 adult female M. muscaedomesticae were added to each box. 10 boxes were used for each treatment. At the end of the 5th day, the mites in each box were separated and counted.

$\frac{ \text{The amount of spent house fly media and the} }{ \text{harvest time} }$

Experiments were done to determine the best amount of media as well as the best harvest time for mass production for this mite. Supplied with 1 ml house fly eggs and 2/3 beaker nematode media for food, 10 adult female M. muscaedomesticae were allowed to propagate 6, 8, or 10 days in 100, 200, or 400 gm of spent house fly media. The number of mites produced in each treatment were compared to select the best size and time combination. A 32-ounce box was used for those treatments with 100 and 200 gm spent house fly media; an 80-ounce box was used for those treatments with 400 gm spent house fly media. Twelve boxes were utilized for each treatment. Previous experimental studies showed that the addition of water was not necessary, and so water was never added.

The number of M. muscaedomesticae

Studies were also performed to determine the proper number of adult female M. muscaedomesticae to be introduced when new boxes were to be tested with 600 gm spent house fly media, 2 ml of house fly eggs, and 2/3 beaker of nematode media added. The box size was 80-ounces and 10, 20, 30, or 40 adult female mites were allowed to propagate in the media for 8 days. At the end of the 8th day, the mites in each box were

separated and counted. Each treatment consisted of 5 boxes with 2 replications, i.e., a total of 10 boxes per treatment.

The amount of house fly eggs

The number of milliliters of house fly eggs to be supplied as food was tested in this experiment.

Eighty-ounce boxes were filled with 600 gm spent house fly media and 1 beaker of nematode media. Then, 0, 2, 4, or 6 ml of house fly eggs were added to the above media. Twenty adult female mites were introduced to each box and allowed to propagate for 8 days. The mites in each box were then separated and counted. Five boxes were used for each treatment with 2 replications, for a total of 10 boxes per treatment.

The amount of nematode media

The quantity of nematode media to be supplied as food was tested in this experiment. The 80-ounce boxes were each filled with 600 gm spent house fly media and 2 ml house fly eggs. Then, 1, 1-1/2, or 2 beakers of nematode media were added to each box. Twenty adult female mites were introduced into each box and allowed to propagate for 8 days. The mites in each box were then separated and counted. Six boxes were used per treatment.

Second test on the effect of house fly eggs

The presence or absence of 10 ml of house fly eggs was tested separately in this study. Six hundred grams of spent house fly media, 20 adult female M.

<u>muscaedomesticae</u>, 1 beaker of nematode media, and the house fly eggs were added to the 80-ounce box. The mites were separated after 3 days and counted. Ten boxes were done for each treatment.

Number of mites per meatballer scoop

A Fairgrove brand meatballer (volume 6.8 cm³) was selected to introduce the M. muscaedomesticae into new boxes. This test was to find out how many mites are ladled in 1 scoop. Two boxes of 7-day-old stock colony were used in this test. The media in the box were stirred to mix the drier top layer with the wetter bottom layer. Ten samples were randomly ladled from each box by the meatballer. The mites in each sample were separated and counted. The sex of this mite can be distinguished in the later deutonymphal stage, so number of female deutonymphs was also recorded.

$\underline{\text{M. muscaedomesticae produced by the meatballer}}$ method

When introducing the mite to a new box with the meatballer, the material (media) ladled by the

meatballer also contains nematodes. This test was done to find out whether there were sufficient nematodes in the new box for the food requirements of the mite. A 32-ounce box was used to measure the spent house fly media. Two boxes of media, one meatballer scoop of the 8-day-old stock colony, and 1 or 0 beaker of the nematode media were added to an 80-ounce box. The mites were separated and counted after 8 days. Eight boxes were maintained for each treatment.

Proportion of adult female M. muscaedomesticae at different days

It was observed that as the colony aged, the sex ratio was changed. Therefore, this study was conducted with M. muscaedomesticae raised by the method previously developed. At the 6th, 7th, 8th, and 9th days, the mites in the box were separated, and the sex ratio (females/total adults) was recorded.

A preliminary study on the improvement of the massproduction method

A preliminary study was done on the effect on reproduction if $\underline{\mathsf{M}}$. $\underline{\mathsf{muscaedomesticae}}$ was fed only with the nematode, $\underline{\mathsf{Protorhabditis}}$ sp.

A colony of M. muscaedomesticae was raised on Protorhabditis sp. (no house fly eggs) in the laboratory for 5 months. The number of offspring produced by this colony was compared with the normal stock colony by the developed method. The 32-ounce boxes were filled with 200 gm of spent house fly media and 1 beaker of nematode media. Then 20 adult female M. muscaedomesticae from this colony or the stock colony were added to each box and allowed to propagate for 5 days. Five boxes were tested for each colony.

The boxes and meatballer used in this study are shown in Appendix 2.

The Modeling Study

Life history of M. muscaedomesticae

Petri-dishes, 9 cm in diameter, were used as ovipositing chambers and those of 5 cm in diameter were used as rearing units. Moistened filter paper was placed on the bottom of the petri-dish to supply moisture and to provide corners for adult female mites to conceal their eggs. Adult male and female mites from the stock colony were introduced into ovipositing chambers and fed with frozen house fly eggs for one day. Then they were transferred to a new ovipositing chamber with fresh frozen fly eggs, 50 pairs per chamber. One hour later, the mites were picked out, and the chambers were searched for mite eggs. The eggs were transferred to the rearing unit, one egg per unit, and were provided with a known number of frozen house fly

eggs. They were then observed at 6-hour intervals. After the mite developed into the adult stage, they were observed once a day and the feeding amount and/or fecundity were recorded. Each female mite was paired with an adult male, which had come from the ovipositing chamber. Rearing units were changed at 2- to 3-day intervals for sanitary purposes.

Generation time in the propagating colony

The mature female deutonymphs and adult males were transferred to an ovipositing chamber. Six hours after this transfer, the newly emerged females and the adult males were transferred again to one-ounce plastic cups that contained spent house fly media and nematode media in a 2:1 ratio, one pair per cup. These cups were examined at 6-hour intervals, 10 cups at each time, for the presence of a second adult female mite--the daughter mite. The experiment was terminated when the daughter mite(s) was found in each of the 10 examined cups. The mean of these 10 observations was calculated to be the mean generation time (T, female to female) of this mite under mass-rearing conditions. Generation times at 4 different temperatures, 22, 26, 30, and 34 C, were studied.

Reproductive rate at different initial population densities

The 32-ounce boxes were filled with 150 gm spent house fly media and 50 gm nematode media. Ten, 20, 40, and 80 adult female mites from the stock colony were introduced to each box. The mites in each box were separated after 72 hours and counted. Each treatment was replicated 4 times.

Results and Discussion

Mass Production of M. muscaedomesticae

The development of a mass-production method for an arthropod species requires certain basic knowledge, including rearing medium, food requirements, development time under specified conditions, number to be produced, and the cost of production.

Studies of Cicolani (1979), Filipponi (1971),
Filipponi and Petrelli (1967), and Filipponi et al.
(1971) showed that the optimum temperature for
population increase of this mite is 30 C. Therefore,
this was the temperature chosen for mass producing the
mites. The incubator used by this researcher did not
have humidity control. Before the experiments, water
pans are placed inside the incubator to raise the
relative humidity to about 60%. During the course of

the experiments, the relative humidity increased to as high as 98%.

The effect of different foods

Based on the study of Rodriguez and Wade (1961), as well as on consideration of the cost of production, spent house fly medium was chosen for mass production of M. muscaedomesticae. Food additives were required in addition to spent house fly medium. The effects of several food additives on the reproduction of this mite were studied, and the results are shown in Table 4-1. All the treatments to which the nematodes, Protorhabditis sp., were supplied as food produced significantly more mites than the rest of the treatments at the 0.05 level in Duncan's multiple range test. Those treatments to which the nematodes were not supplied were not significantly different from the check. Two conclusions can be deduced from this result. First, the nematode is the principal food for the propagation of this mite. Second, the other foods did not improve the reproductive rate of this mite. The effect of the house fly eggs and the nematodes on the reproduction of this mite has been studied by Ito (1973) and Rodriguez et al. (1962). In order to compare this research with their studies, the reproductive rate (progeny/female/day) was calculated as: number of mites produced divided by (5 (days) x 10

Table 4-1. The effects of different foods, either singly or combined, in spent house fly medium, on the reproductive rate of $\underline{\text{M}}$. $\underline{\text{muscaedomesticae}}$ at 30 $^{\pm}$ 2 C, 60-98% R. H.

Food*	No. mites a	fter 5 days	Dans/61-/4	
	X**	SE	Progeny/female/day	
E+N+F	408.7a	65.9	8.17	
E · M · E	400.74	03.9	0.17	
E+N	444.2a	53.3	8.88	
E+F	171.2b	9.0	3.42	
N+F	433.6a	53.4	8.67	
E	115.6b	12.6	2.31	
N	443.1a	67.1	8.86	
F	114.86	21.1	2.30	
control	79.0b	16.8	1.58	

^{*} E: Frozen house fly eggs, 1 ml. N: Nematode medium, 2/3 beaker.

F: 0.01% solution of fish emulsion fertilizer. 30 ml + commercial layer feed, 10 gm.

^{**} Mean of 10 boxes. Means followed by the same letter were not significantly different at the 0.05 level in Duncan's multiple range test.

(female mites)). Both of their studies agreed with this study on the importance of nematodes. The presence of nematodes always produced higher numbers of this mite. The experiment in the modeling study of this chapter also demonstrates that this mite species has a higher reproductive rate when fed with nematodes, However, the effect of house fly eggs was not consistent among these 3 studies. In the study of Rodriquez et al., the highest number of progeny was produced by supplying both nematodes and house fly eggs as food, followed by supplying nematodes only, then by supplying house fly eggs only. The reproductive rate was higher in their study than in this study. This may be because the food was added daily in their study. Further studies of Ito (1973, 1977b) also showed that the daily addition of food, the house fly eggs and/or the nematodes, can improve the reproduction of this mite. This alternative was not considered by this researcher because the method would require more labor and thus increase the cost of mass production. The addition of fish meal and soybean oil meal to the spent house fly media has been reported to produce more mites (Rodriguez and Wade 1961). In this study, the addition of fish emulsion fertilizer and commercial layer feed did not produce significantly more mites than the check. The addition of the nematode produced much higher numbers of M. muscaedomesticae whether these additives were included

or not. Therefore, it was concluded that fish emulsion fertilizer and commercial layer feed should be omitted to lower the cost. The house fly eggs, although shown to be ineffective, were still added for food to preclude the possible change of the food habits of this mite in the mass-production colony.

The nematode, Protorhabditis sp. (as determined by Dr. A. C. Tarjan), is a new species of nematode that is found to be the prey of M. muscaedomesticae. It was first noticed in the laboratory colony of M. muscaedomesticae. Tests on the spent house fly media. frozen or unfrozen, did not find these nematodes. Observations of the mites indicated that mites can carry the nematodes (Figure 4-1). Therefore, this nematode may have come from caged-hen manure, collected along with the mite. The nematodes hide in the coxal cavities, gnathosomal cavity, peritremes, and wrinkles on the body surface of the mite (observed in that order of frequency). With a binocular microscope, one can observe the nematodes struggling out from their hiding places on the mite when it is immersed in water or alcohol. Both male and female mites can carry nematodes. Among the observed mites, 100% of starved mites carried the nematodes. Only a few well-fed mites carried the nematodes. A number of mites were separated from the substratum and kept in a petri-dish without any supply of food or medium to study how long a mite



Figure 4-1. $\underline{\text{M}}$. muscaedomesticae carrying nematodes, $\underline{\text{Protorhabditis}}$ sp.

will carry a nematode. The nematodes were seen to flow away from the coxal cavities of the mites after 12 days. Poinar (1965) reported that individuals of Macrocheles glaber and M. submotus carry a nematode (Pelodera acarambates), a possible food for them, on their back. Carrying nematode prey may be a general phenomenon among the manure-inhabiting macrochelids. The rhabditid nematodes almost invariably carry with them the bacteria that they feed. It appears that every member in this food chain carries its own food. Therefore, the macrochelid mites will not have problems with an adequate alternative food supply as usually happens with predators.

Mass production of the nematodes had been studied by Singer and Krantz (1967) and Singh and Rodriguez (1966). The method presented here is a more economic and convenient method for mass production for these mites.

The amount of spent house fly media and the harvest time

After testing the value of various foods, the next study was to determine the quantity of spent house fly media to be used in a rearing unit and the proper culture time before the harvest. The result of a factorial comparison of the effects of 3 amounts of spent house fly media and 3 different time periods on

the reproduction of M. muscaedomesticae is shown in Table 4-2. The analysis of variance showed both factors had a significant effect on the reproduction of this mite. For each factor there were significant differences between levels (different amounts of spent house fly media or days). In addition, there was an interaction effect between these 2 factors. Duncan's multiple range test was made on all treatments to find out the best combination of the 2 factors. A significantly higher number of M. muscaedomesticae were produced when 400 gm of spent house fly media were used to propagate the mite for 8 days. Theoretically, a population will increase exponentially when there is no environmental restriction. With 100 gm or more of spent house fly media, the population of this mite increased exponentially within 6 days. After the 6th day, neither 100 gm nor 200 gm of spent house fly media were optimal for the reproduction of the mite. The mite population in 100 gm of spent house fly media decreased after the 6th day, though not statistically significant. Six days appeared to be the optimal time period for the treatment of 100 gm of spent house fly media. The rate of population increase of this mite slowed down after the 6th day in the 200 gm spent house fly media. although their total number was increasing. Only in 400 gm of spent house fly media did the exponential

Table 4-2. Population of $\underline{\text{M}}$. muscaedomesticae as affected by quantity of spent house fly medium at different time intervals at 30 $^{\pm}$ 2 C, 60-98% R.H.

Quantity medium		of	No. mites after x days				
	m)		6	8	10		
100	X SE		927.5bc 98.6	796.7ab 13.0	325.7a 74.9		
200	X SE		820.0abc 85.9	1394.4c 144.9	950.0bc 118.8		
400	X SE		963.3bc 128.2	3706.7e 322.4	2498.3d 346.6		

^{*} Mean of 12 boxes. Means followed by the same letter were nou0significantly different at the 0.05 level in Duncan's multiple range test.

increase of the mite population continue until the 8th day. After the 8th day, the mite population decreased.

The actual role of the amount of spent house fly media may have been a function of the rate of nematode multiplication in the substratum. The greater amount of media provided more nematodes as food for M. muscaedomesticae. The nematodes have been shown to be valuable in reproduction of this mite in the previous experiments on the effects of different foods. On the other hand, the multiplication of the nematodes can make the spent house fly media wet, making it unsuitable for production of M. muscaedomesticae. In this experiment, the spent house fly media was quite wet after 6, 8, and 10 days for the treatment of 100, 200, and 400 gm of media, respectively. Rodriguez et al. (1962) "pre-seeded" the media with nematodes 2 days prior to the introduction of M. muscaedomesticae. These treatments all produced a lower number of mites in comparison with those treatments that inoculated nematodes at the same time as the mites. This probably was the result of the wetting of the media by the nematodes.

After this experiment it was realized that the 80-ounce box, which could hold more media, was best suited for rearing. Therefore, 600 gm of spent house fly media were used in the following experiments. Based on this experiment, 8 days was chosen to be the best

time period for the development of the mass-production method.

The number of M. muscaedomesticae

After deciding the amount of spent house fly media to be added to each box and the time period for the propagation of this mite, the next question was the number of mites that should be introduced into each box. The number of progeny of 10, 20, 30, and 40 adult female M. muscaedomesticae were studied and the results are shown in Table 4-3. Introduction of 20 adult female mites to a new box gave the best results. Introducing more than 20 adult female mites did not produce significantly more progeny, and a crowding effect can be seen from the decreased number of mites produced. The introduction of 20 adult female mites was chosen for the following experiments as this number produced a significantly higher number of offspring than introducing 10 adult female mites, and provided more information on the number of mites that could be produced.

The amount of house fly eggs

The effect of the amount of house fly eggs to be added to a new box for the food of $\underline{\mathsf{M}}$. $\underline{\mathsf{muscaedomesticae}}$ is shown in Table 4-4. The numbers of mites produced in

Table 4-3. Progeny of $\underline{\text{M}}$. $\underline{\text{muscaedoesticae}}$ after 8 days based on original number of adult female mites introduced, at 30 $^{\pm}$ 2 C, 60-98% R.H.

No. adult females	No. mites a	fter 8 days
introduced	<u>X</u> *	SE
10	2390a	245.2
20	3664b	317.6
30	3146ab	223.0
40	3086ab	291.4

^{*} Mean of 10 boxes. Means followed by the same letter were not significantly different at the 0.05 level in Duncan's multiple range test.

Table 4-4. Populations of \underline{M} . \underline{M} muscaedomesticae and the effect of house fly eggs at 30 \pm 2 C, 60-98% R.H.

Quantity of house	No. mite af	ter 8 days
fly eggs (ml)	<u>x</u> *	SE
6	2618.9a	245.7
4	2845.7a	117.3
2	2854.3a	173.5
0	2846.0a	257.4

^{*} See Table 4-3.

all 4 treatments were not significantly different from one another at the 0.05 level in a Duncan's multiple range test. Nematodes had also been added as food in each treatment. This experiment again showed that the supply of house fly eggs in the presence of nematodes did not improve reproduction of this mite. However, the addition of 2 ml of house fly eggs to a new box was selected for the following experiments.

The amount of nematode media

The effect of the amount of nematodes (nematode media) on the reproduction of $\underline{\mathsf{M}}$. $\underline{\mathsf{muscaedomesticae}}$ in spent house fly media is shown in Table 4-5. The mite population produced by the addition of the 3 tested amounts of nematode media was not significantly different at the 0.05 level in Duncan's multiple range test. Based on the results of this study, 1 beaker of nematode media was added as the food for mass production of $\underline{\mathsf{M}}$. $\underline{\mathsf{muscaedomesticae}}$.

Second test on the effect of house fly eggs

Advanced studies were made after the initial series of experiments. A more economical method could be achieved by maintaining a stock mite colony and propagating this mite from the stock colony for biological control or research. The stock colony would

Table 4-5. The effect of nematode, Protorhabditis sp., as a food source on the population of M. muscaedomesticae after 8 days, at 30 $^{\pm}$ 2 C, 60-98% R.H.

Quantity of nematode	No. mites after	-
medium (Beaker)	<u>x</u> *	SE
2.0	3473a	296.7
1.5	4247a	309.1
1.0	4417a	365.3

^{*} Mean of 6 boxes. Means followed by the same letter were not significantly different at the 0.05 level in Duncan's multiple range test.

be supplied with both house fly eggs and nematodes as food. The colony to be propagated (propagating colony) from the stock colony would be supplied only with nematodes as food. This method has the following advantages over the previous one. 1. More house fly eggs can be supplied to the stock colony, yet the total requirement of house fly eggs is fewer than before. 2. A change in food habits of the insectary-produced M. muscaedomesticae will be less likely. This mite can produce approximately 2-1/2 generations in 8 days, which should not be long enough to induce changes in food habits in the propagating colony. Sufficient house fly eggs are supplied to the stock colony so that the stock colony will not change its food habits either. 3. The cost will be lower. Ten milliliters of house fly eggs was selected as the amount to be added to the stock colony. The effect of 10 ml of house fly eggs was compared with no house fly eggs in Table 4-6. The results were as expected. There was not a significant difference between the 2 treatments.

Number of mites per meatballer scoop

In a mass-production technique, introduction of mites into new boxes by counting their number is not realistic. To simplify the prodedure, a meatballer was selected for introducing M. muscaedomesticae from the stock colony to the propagating colony. A test was

Table 4-6. Effect of extra amounts of house fly eggs on the population of M. $\frac{\text{muscaedomesticae}}{\text{medium, at 30 }^{\pm} \text{ 2 C, }} \frac{\text{muscaedomesticae}}{\text{60-98\$ R.H.}} \text{ in spent house}$

Quantity of house	No. mites	after 8 days
fly eggs (ml)	<u>x</u> *	SE
10	2867.6a	131.6
0	2501.7a	102.9

^{*} See Table 4-3.

conducted to determine the number of mites that one took from the stock colony per meatballer scoop. The results are shown in Table 4-7. One is able to distinguish the sex of the mature deutonymph of this mite by the shape. At 30 C, the deutonymphs will emerge to adult within half a day or one day. Therefore, the number of female deutonymphs was recorded and considered as adults. The distribution of this mite in the meatballer samples was uniform, with standard errors less than 10% of the mean. The sum of female adults and female deutonymphs, 34.5, is slightly higher but still acceptable (see Table 4-3). Samples of smaller quantity of stock colony media had a larger standard error with the ratio of $S_{\overline{v}}/\overline{X}$ larger than 0.10. A uniform number of female mites per meatballer is important. Therefore, this meatballer was accepted for introducing mites. However, the presence of male mites ensured the fertilization of female mites, thus accelerating the propagation of the mite.

$\underline{\text{M. muscaedomesticae produced by the meatballer}}$ $\underline{\text{method}}$

Previous testing demonstrated that a uniform number of female mites could be introduced with a meatballer. The next step was to test what mite population would be produced with this method. The studies also showed that the nematode (Protorhabditis

Table 4-7. Number of M. $\underline{\text{muscaedomesticae}}$ in one scoopful of 8-day-old stock colony $\underline{\text{media.}}$

	Female	DN*	Subtotal	Male	Nymph	Total
<u>X</u> **	24.65	8.85	34.50	14.70	19.65	68.85
SE	2.27	0.77	2.43	1.17	1.40	4.17
$S_{\overline{X}}/\overline{X}$	0.092	0.087	0.091	0.079	0.071	0.061

^{*} The mature deutonymph which will emerge to adult female soonly.

^{**} Mean of 20 scoops.

sp.) multiplied on the spent house fly media as did the M. muscaedomesticae. The material (medium) ladled by the meatballer contained not only mites but also nematodes. Would there be sufficient nematodes to multiply and serve as food for the mite population so that no nematode colony would be required? This was tested and the results are shown in Table 4-8. The nematodes in one meatballer of the 8-day-old stock colony media were too few to multiply sufficiently as food for the mite population. Without adding additional nematodes, the mite population produced was significantly smaller. Therefore, it was necessary to add 1 beaker of nematode media.

By using the meatballer, the number of mites produced was not different statistically from most of those of the previous experiments. This technique could be a breakthrough because it saves labor and time. Without this approach, the developed method cannot be considered as a mass-production method.

Proportion of adult female M. muscaedomesticae at different days

Filipponi (1964), Filipponi and Petrelli (1967), and Filipponi et al. (1971) reported that in the laboratory-reared population of M. muscaedomesticae, the sex ratio (females/total adults) oscillated. The variation in the sex ratio was also noticed by this

Table 4-8. M. musçaedomesticae produced in spent house fly medium by different rearing techniques at $30\ ^-$ 2 C, 60-988 R.H.

	Oua	Quantity of		N	No. mites after	fter	
Mite	Spent house	House flv	Nematode		8 days		SX/X
1 1 1 1 1 1 1 1 1	fly medium	eggs (ml)	medium (beaker)	z	*	SE	1
		T	This experiment				
l scoop	2 boxes of 32 ounce box	0	1	∞	2270.0c	77.6	0.038
=	=	0	0	œ	1755,5d	6*68	0.051
		Pre	Previous experiments				
20 F**	mb 009	0	2/3	10	2846.0c	257.4	0.090
20 F	mb 009	1	2/3	10	3664.0b	317.6	0.087
20 F	mb 009	2	2/3	10	2854.3c	173.4	0.061
20 F	mb 009	0	1	10	2501.7c	102.9	0.041
20 F	mb 009	2	1	9	4417.0a	365,3	0.083
20 F	mg 009	10	1	10	2867.60	131.6	0.046
* Means Duncan	Means followed by the same le Duncan's multiple range test.	same letter v	* Means followed by the same letter were not significantly different at the 0.05 level in Duncan's multiple range test.	differ	ent at th	e 0.05	evel in

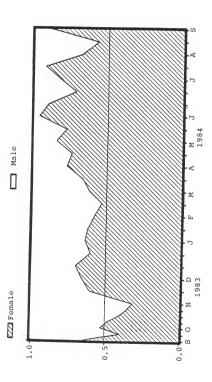
^{**} Female adults.

researcher. As the adult female ate much more than the other stages of this mite (Table 4-13), it would be more important to produce female mites. It should be noted that this is an arrhenotokous species and that only fertilized eggs produced females. Therefore, the sex ratios in the propagating colony at different time periods were studied to determine when (culture time) the most female mites are produced. The results are shown in Table 4-9. The proportion of the adult female mites (the sex ratio and the ratio of adult females to total population) increased as the colony aged. The propagating colony contained only adult female mites after 11-12 days. In the studies of Filipponi and Petrelli (1967) and Filipponi et al. (1971), the sex ratio averaged 0.758 in the population and 0.440 at birth. They increased the amount of medium and food progressively. Therefore the mite population in their study was not under environmental restrictions. The sex ratios that they observed were probably the same as the sex ratios of this mite when there is unlimited reproduction. In this study, the sex ratio at the 6th day was closer to the sex ratio at birth as indicated by Filipponi and Petrelli and Filipponi et al.; and the sex ratio at the 9th day was closer to their population sex ratio. The sex ratio in the field population is drawn in Figure 4-2. This shows that the sex ratio fluctuated in the poultry manure, being lowest on

Table 4-9. The proportion of the adult female mites in the mass-production population of M. muscaedomesticae at different days at 30 $^\pm$ 2 C, 60-98% R.H.*

Days after set up	Ratio of females/total adults	Ratio of females/total population
6	0.5355a	0.4298a
7	0.5853ab	0.5015ab
8	0.6252b	0.5324b
9	0.7317c	0.6661c

^{*} See Table 4-3.



Sex ratio of M. muscaedomesticae in caged-hen manure (mean of Figure 4-2. 20 samples).

November 4, 1983 (0.312), and highest on June 1, 1984 (0.965). This would appear to indicate an environmental effect. The study by Ito (1977b) also showed that the sex ratio in the colony of the mite species changed as the colony aged. Cicolani and Bullini (1975) studied the sex ratio of Macrocheles matrius. A similar change in sex ratio existed in their results.

Though the proportion of the adult female $\underline{\mathsf{M}}$. $\underline{\mathsf{muscaedomesticae}}$ in the population at the 9th day was significantly higher than at the 8th day, the 8th day was preferred by this researcher to be the harvest day for the following reasons:

- 1. The multiplication of the nematodes was destroying the substratum. At the 8th day, the quality of the substratum is always good, but may not be at the 9th day.
- Food (in the form of nematodes) was greatly reduced after the 8th day.
- 3. The increase in the sex ratio resulted from the decrease in the number of males, young, and total population numbers. This was clearly shown in the study of Ito (1977b). This researcher found that after starvation, the female mites tend to produce male offspring at first and then, later, may produce female offspring. Filipponi and Petrelli (1967) observed both sexes of this mite were capable of mating more than once. The number of fectilized eggs laid after one

mating was small and the sex ratio at birth was negatively correlated to the population sex ratio. Their studies suggested that this mite needs to mate often to keep on laying fertilized eggs. Thus the lack of adult male mites will delay the population increase of this mite after field release.

- 4. As the total numbers decrease, the actual number of female mites on the 9th day may not be greater than on the 8th day.
- 5. The laboratory colony of this mite was often contaminated by an ascid mite. In such cases the population of the ascid mite was much lower at the 8th day.
- 6. There is a time lag between the harvest and release. This would cause the total time period to be longer than 8 days. At the time of application, the proportion of adult females will be closer to 0.758.

Therefore, a primitive method for mass-producing M. <u>muscaedomesticae</u> was developed. In summation, it is as follows:

Mite colony

A. Stock colony

Container: the Superseal brand 80-ounce oblong food saver.

Substratum:

 Spent house fly media: 2 boxes, measured by the Superseal brand 32-ounce oblong food saver.

- 2. Frozen house fly eggs: 10 ml.
- Nematode media: 1 beaker, 3-5 days old, measured by the 50 ml tri-pour disposable beaker made by Sherwood Medical Industries Inc.
- Amount of M. muscaedomesticae to introduce:

 1 ladle of 8-day-old stock colony media,
 measured by the Fairgrove brand meatballer
- Time period for the mite to propagate before harvest: 8 days.
- B. Propagating colony: the same as the stock colony except it does not have the supply of frozen house fly eggs.

Nematode colony

Container: widemouthed quart glass mason jar.

Substratum: spent house fly media; fill up to 4

cm beneath the mouth.

Amount of nematodes to inoculate: 1-2 tablespoonfuls of 3- to 5-day-old nematode colony.

Time period for nematodes to multiply before use: 3-5 days.

The cost of raising 500,000 M. muscaedomesticae daily, 5 days a week, was estimated in Table 4-10.

Assuming 2500 mites are produced per box of propagating colony, 200 boxes will be required to produce 500,000

Table 4-10. Cost of producing 500,000 $\underline{\text{M}}$. $\underline{\text{muscaedomesticae}}$ daily, 5 days a week.

Items	Cost (\$)	Annual Cost (\$)
Initial cost	7410.00	842.00
Building (20' x 20' concrete block)	6400.00	640.00
Freezer	450.00	90.00
Air conditioner	450.00	90.00
Humidifier	110.00	22.00
Materials for producing 500,000 mites on daily basis	758.00	
Shelves	215.00	
Superseal oblong food saver		
80 ounce (x 230)	476.10	
32 ounce	2.92	
Widemouth quart Mason jars (x 44)	33.00	
Tri-pour 50 ml beaker	3.36	
Meatballer	2.50	
C-299 maiden chiffon (9 yards)	25.12	
Maintenance (for producing 500,000 mites)	81.10	
Labors (2 men, 200.00/man/week)	80.00	
Utilities (water, electricity, 22.00/month	1.10	

N.B. No cost given for media as spent house fly media was provided free-of-charge from USDA laboratory, Gaines-ville, Florida.

mites. The mites in one stock colony box can inoculate 30 boxes of propagating colony. Seven boxes of stock colony are needed to inoculate those 200 boxes of propagating colony, and one more box is needed to maintain the stock colony itself. Therefore, 208 boxes of mites will be raised. Approximately 35 mason jars of nematode media are needed to provide food (nematodes) for the 208 boxes of mites. An additional 5 jars are needed to maintain the nematode colony. An extra 10% additional boxes and jars are recommanded in case of breakage. Two full-time working employees are required to take care of both mite and nematode colonies. Spent media from rearing 3 trays of house flies can fill approximately 13 boxes. A house fly colony of 52 trays will provide spent media for raising 500,000 mites (48 and 4 trays for the mite and nematode colonies, respectively). An estimated 11.7 and 6 trays, respectively, are needed to manintain the fly colony and to provide food (fly eggs) for mite stock colony (see Morgan 1981a,b). The remaining 34.3 (63.52%) trays of house fly colony are merely for acquiring spent medium. It is too costly if one does not produce these mites in association with house fly production. Therefore, the cost of house fly production is not included in Table 4-10. The cost may be high initially, but the maintenance cost is inexpensive.

Possible problems with this method

During the study of the mass-production of \underline{M} . muscaedomesticae, there were 2 problems:

First, a strong odor of ammonia was always generated in the incubator when more than 10 boxes of these mites were propagated in the incubator. According to Wallwork and Rodriguez (1963), the ammonia had an adverse effect on the reproduction of this mite when it reached a certain concentration. The presence of ammonia may partially account for the lower reproductive rate of this mite in the later experiments. Therefore, good ventilation must be maintained in the insectary to prevent ammonia concentration in the air.

Second, as previously stated, an ascid mite often contaminated the <u>M. muscaedomesticae</u> colony. It can invade the nematode colony too. The propagation of this ascid mite in the substratum resulted in a lower number of <u>M. muscaedomesticae</u> (by as much as 100-200) produced per box. When this ascid mite contaminates the nematode colony, the problem can be solved easily by adding water to the nematode colony and then filtering with C-299 maiden chiffon, which will filter out only the nematodes, and then a nematode colony can be re-established. The new nematode colony is available for use after 3 days--without any major time delay. However, when the <u>M. muscaedomesticae</u> colony is

contaminated by this ascid mite, there is no easy way to eliminate it. The ascid mites were observed frequently to adhere on the dorsum of adult female M. muscaedomesticae and disperse along with M. muscaedomesticae. Checking the M. muscaedomesticae individually under a binocular microscope and re-establishing a new colony from these clean M. muscaedomesticae is the only way to eliminate this ascid mite. Acarid mites were reported to be phoretic on macrochelid mites (Axtell 1969 and Chant 1960). Other smaller species of mites have been seen attached to M. muscaedomesticae in caged-hen manure. The control of these ascid mites should be studied for the following reasons: 1. To prevent the initial contamination of the mass-production colony of M. muscaedomesticae. 2. To find simple ways to eliminate them after contamination. 3. To reduce the indigenous field population of these mites to increase the efficiency of M. muscaedomesticae. This may be achieved by chemical control. In the study in Chapter 5, it was observed that these ascid mites died at concentrations that had little effect on M. muscaedomesticae. Before there are better methods to control the ascid mites, 2 rules should be followed in the insectary. l. Always cover the M. muscaedomesticae colony with the lid of the box and the nematode colony with the C-299 maiden chiffon. This will reduce the chances of contamination.

2. Do not keep any colony after it is 9 days old. Clean them out on the 9th day. Laboratory contamination by this ascid mite always happened after neglect of sanitary conditions.

Future studies to improve this method

$$N_t = N_o e^{rT}$$

where r = 1n $\rm R_{\rm O}/T$ = 1n (58.07)/4.4845 = 0.905708. So if we start with 10 female mites, we will have 10 x exp(0.905708 x 8) = 14,020 mites after 8 days. However, studies on the reproductive rate of this mite species at different population densities have shown that the

reproductive rate r decreased when the population density increased. Based on one study which inoculated 160 adult female mites per propagating colony box and produced an average of 4006.5 and 4084.0 mites, respectively, at the 3rd and 5th days, the carrying capacity of this mass-production method was approximately 4500 mites per box. It would not be unreasonable to expect a production routinely of 3000 to 4000 mites per box.

Improvement of this method may be accomplished in the following manner:

1. As previously discussed, the reproductive rate of M. muscaedomesticae in the substratum used in this method ranged from 15.64 to 46.33. The ratio of standard error over mean $(S_{\overline{X}}/\overline{X})$ in these experiments was always smaller than 0.10, with many smaller than 0.05. Some of the ratios are shown in Table 4-8. The variation in the reproductive rate of this mite species was probably the result of the variation in the quality of spent house fly media and the biotic variation among individual mites. Therefore, the standardization of the quality of the spent house fly media should be studied. Moisture content may be an important character for the spent house fly media. Rodriguez and Wade (1961) and Rodriguez et al. (1962) oven-dried the spent house fly media before use, then mixed it with the 5 N water solution of sodium hydroxide in a certain ratio. This

may be a way to standardize the quality of the spent house fly media. Another possible method to standardize the quality of media is to use new instead of spent house fly media. The study of Rodriguez et al. (1962) showed reproductive rates for M. muscaedomesticae of 17.5 progeny per female per day can be achieved in new CSMA house fly media. With this reproductive rate, the inoculation of 20 adult female mites can produce 2800 mites after 8 days. The success of this method will greatly reduce the dependence of mass production of M. muscaedomesticae on the production of the house fly, and thus reduce the cost. The major problem in using the new house fly media may be how to make it suitable for the multiplication of the nematode, Protorhabditis sp., or other nematodes that are nutritious to M. muscaedomesticae.

2. Studies of Ito (1973) and Rodriguez et al. (1962) showed that by adding food daily, a larger number of M. muscaedomesticae can be produced on a smaller quantity of substrata. However, the daily addition of food requires too much labor time. A compromise method would be to fill the box with half the amount of the substratum and then add the remaining half at the 5th day. The nematodes multiply in the spent house fly media at a rate faster than the mites. The population of nematodes decreased after peaking in the container. The spent house fly media at this time had a lower

quality for the reproduction of both the mite and the nematode. By adding half the amount of the substratum later, a better environment may result on the 5th to 8th day and thus increase the production of $\underline{\mathsf{M}}$. muscaedomesticae.

- 3. There were 34.5 female adults and mature deutonymphs in one scoopful of 8-day-old stock colony media, which was more than what was needed. If a better simpler, technique that would introduce 15-20 female mites could be found, it would reduce the cost and give better production. Introduction of a larger number of female mites may serve as a method to mass produce this mite within a shorter time period in the case of the emergence of large numbers of house flies.
- 4. The optimal oviposition temperature for $\underline{\mathsf{M}}$. $\underline{\mathsf{muscaedomesticae}}$ ranged from 28 C to 36 C (Filipponi and Petrelli 1967, Filipponi et al. 1971). A comparison of the cost of mass producing this mite at a temperature lower than 30 C should be done to see if it is more economical.
- 5. The nematode, <u>Protorhabditis</u> sp., has a higher nutrient value than other foods in the reproduction of <u>M. muscaedomesticae</u>. When supplied with this nematode, additional foods did not increase the numbers of the mite produced. This nematode is probably a natural food of <u>M. muscaedomesticae</u> and is very easy to mass produce in the laboratory. The cost of the mass production of

- M. muscaedomesticae would be greatly decreased if frozen house fly eggs could be omitted from the food list of this mite. A preliminary study on this has been done. A colony of M. muscaedomesticae fed only with nematodes was cultured in the laboratory for 5 months. Then the reproduction of this colony was compared with the stock colony. The results are shown in Table 4-11. There were significant differences between the 2 colonies in the number of progeny produced. However, the stage composition was not different. Therefore, feeding only the nematodes seems to have no adverse effect on the reproduction of this mite. However, the long-term effect on the food habits and predation rate of this mite on house fly eggs and 1st-instar larvae should be studied before omitting frozen house fly eggs from the food list of the mite. If it can be done, the combination of this and the use of new house fly media will make the mass production of M. muscaedomesticae independent from house fly rearing.
- 6. The best future for the mass production of M.

 <u>muscaedomesticae</u> is the joint mass production of the
 mite and the parasites of the house fly, e.g. <u>Spalangia</u>
 <u>endius</u>. M. <u>muscaedomesticae</u> would be integrated with
 other agents to control the house fly. The house fly
 pupal parasite, <u>Spalangia</u> <u>endius</u>, has controlled
 poultry house flies effectively (Morgan et al. 1975)
 and can be mass produced (Morgan 1981b) and marketed

Table 4-11. Effects of feeding nematodes and house fly eggs compared to nematodes only on the number of female M. muscaedomesticae after 5 days, at 30 ± 2 C, 60-98% R.H.*

		NE**	N**
mites			
Females		121.6a	192.4b
Total		342.8a	514.8b
os			
Females/total	adults	0.591a	0.645a
Females/total	population	0.358a	0.373a
E	Females Total Os Females/total	remales Total	nites Pemales 121.6a Potal 342.8a ps Pemales/total adults 0.591a

^{*} Mean of 5 boxes. Means in the same horizontal line followed by the same letter were not significantly different at the 0.05 level in Duncan's multiple range test.

^{**} NE: The laboratory strain which was supplied with both nematodes and frozen house fly eggs as food.

N: The laboratory strain which was supplied with

nematodes only as food. Both strains had been maintained in the laboratory for 5 months before this test.

for poultry house fly control. As previously discussed, when mass producing \underline{M} . $\underline{\text{muscaedomesticae}}$, only a small portion of the produced house flies were needed for laying eggs to maintain the house fly colony and to supply food for the mite colony; the majority were just for acquiring the spent house fly media. The joint mass production of both parasites and mites can utilize all the materials of the system, and thus, nothing is wasted.

The production of both the most effective parasite and a predator of flies in poultry houses as is presently known can increase the benefits. Referring to the report of Morgan (1981a,b), approximately 479,768 Spalangia endius can be produced together with every 500,000 mites. The calculations are as follows. The portion for acquiring spent media is 63.52% or 34.3 trays of house fly colony. This can produce 686,000 house fly pupae. For every 100 parasitized pupae, 80 Sendius are produced. Therefore, 548,800 parasitoid wasps can be produced from 686,000 house fly pupae.

Among the wasps, 69,030 will be needed for parasitizing fly pupae to produce more wasps, and 479,768 wasps are available for field release.

7. The incubator used in this study did not have humidity control. During the experiments, the relative humidity ranged from 60% to 98%, which is not ideal for the multiplication of this mite. Maintaining the

relative humidity in the range of 70-80% would result in better production.

In addition, the field test in the study of the evaluation of this mite showed that it is necessary to improve the productivity per unit box, and therefore, decrease the amount of media to handle in field release of the mites.

In order to obtain quality control of the produced mites, the following should be done to increase the gene pool of the insectary population:

- Several strains of this mite collected from different areas should be maintained in the insectary.
 The stock colony should be interbred with these strains.
- Adult male mites collected from the poultry farm should be added to the stock colony periodically.

The Modeling Study

Initially, a simplified model was hypothesized for this species of macrochelid mite, which would allow for accurate prediction of harvest time. This involved a series of experiments that determined stage mortality, sex ratio, generation time, and the change of reproductive rate when the population density was increasing at a constant temperature and relative humidity.

Life history of M. muscaedomesticae

The rapid development time of the mite along with its natural habit of remaining within the substrate made it almost impossible to observe its life history in spent house fly media. The color and the number of hiding places in this type of media make the mites invisible. Therefore, petri-dishes were used for this study.

Life cycle data of M. muscaedomesticae is shown in Table 4-12. The developmental time from egg to adult and adult longevity were 66.5 hours and 21.6 days for male mites, and 70.0 hours and 21.9 days for female mites, respectively. These figures differ from other authors apparently because of diet. The sex ratio was 0.625. Table 4-13 shows the number of house fly eggs fed on by this mite. The number of eggs fed on by mites may be affected by the decay of the eggs, which occurs at 30 C. The larval stage did not feed. The protonymphal stage ate few, while the deutonymphal stage consumed many. Female nymphs ate almost twice as many as male nymphs. The feeding amount of adult female mites shown in Table 4-13 included those of their mates. Adult male mites, observed separately, consumed approximately 1.6 fly eggs per day per mite. This was a small portion of the amount of the fly eggs that were consumed by adult female mites, especially during the

Table 4-12. The life cycle (hours) and longevity (days) of M. muscaedomesticae when fed with frozen house fly eggs at 30 $^{+}$ 2 C.

	м *	r*	*Nd	* NO	Total	Adult** longevity	Sex ratio (females total adults)
Female (n=39)		1					
×	15.54	0.9	21,38	15.54 6.0 21.38 27.08 70.0	70.07	21.9	
SE	0.85	0.0	0.58	0.85 0.0 0.58 1.02	1,39	2.18	
							0.625
Male (n=25)							
I×	16.8	0.9	21.84	16.8 6.0 21.84 21.84 66.48	66.48	21.63	
SE	1.04	0.0	0.58	1.04 0.0 0.58 1.24 1.54	1.54	1,89	
* E: egg, L: larva, PN: protonymph, DN: deutonymph.	larva	, PN:	L: larva, PN: proton	Amph, D	mph, DN: deuto	onymph.	
** $n=20$ for female mites, $n=16$ for male mites.	Eemale	mites	, n=16	for mal	e mites		

Table 4-13. Feeding amount of $\underline{\text{M}}\text{.}$ $\underline{\text{muscaedomesticae}}$ on frozen house fly eggs at 30 $^{\pm}$ $\overline{2}$ C.

Stages*	Mal	e**	Fema	le***
o cayes.	x	SE	X	SE
L	0.0		0.0	
PN	0.7	0.2	1.5	0.3
DN	7.5	1.0	13.5	0.9
Subtotal	8.2	0.9	15.0	0.9
Adult****				
Oviposition period			221.0	24.8
Total no. consumed as adult	34.3	3.5	247.2	24.3

^{*} See Table 4-12.

^{**} n=10 for immature stages, n=16 for adult stages.

^{***} n=22 for immature stages, n=20 for adult stages.

^{****} Female consumption includes amount consumed by their mates.

ovipositing period. These data again indicate what an excellent predator of house flies this species is.

The ovipositing period and fecundity of $\underline{\mathsf{M}}$. <u>muscaedomesticae</u> are shown in Table 4-14. Figure 4-3 shows the average fecundity curve for 20 female mites with the egg laying day indicated. The maximum number of eggs laid per day occurs from the 11th to the 13th day after eclosion. The weighted average egg laying day is about 10.14 $^{\pm}$ 92 days after adult eclosion and is indicated in Figure 4-3 by the vertical line. The stage mortality is diagrammed in Figure 4-4. Figure 4-5 gives the initial concepts on life history as determined by the above experiments.

Generation time in propagating colony

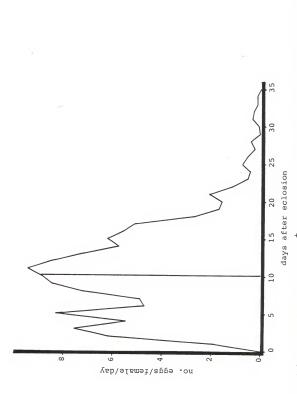
The generation times of M. muscaedomesticae when reared in spent house fly media with the nematodes, i.e., in a propagating colony, at 4 different temperatures are shown in Table 4-15.

Reproductive rate at different initial population densities

The reproductive rates of \underline{M} . \underline{M} \underline

Table 4-14. The pre-, post-, and ovipositing period (days) and fecundity of $\underline{\text{M}}$. muscaedomesticae when fed with frozen house fly eggs at 30 $^{\pm}$ 2 C, n=20.

	\overline{x}	SE
Preovipositing period	2.2	0.67
Ovipositing period	15.8	1.65
Postovipositing period	3.8	0.99
Eggs/female	124.9	15.86
Eggs/female/day	8.05	0.99



time. Fecundity curve at 30 $^{\pm}$ 2 C from eclosion to end of life averaged for muscaedomesticae. Vertical line is weighted average reproductive time Figure 4-3. 20 female \underline{M} .

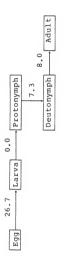


Figure 4-4. Life history and stage mortality of $\underline{\mathrm{M}}$. muscaedomesticae.

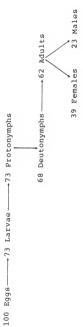


Figure 4-5. Idealized life history for 100 eggs of $\underline{\mathrm{M}}_{\cdot}$ muscaedomesticae.

Table 4-15. Generation time (days) of M. muscaedomesticae feed on the nematode, Protorhabditis sp., in spent house fly media at 4 different temperatures and 60-98% R.H. with 10 female mites per observation.

Temperature	Generation t	ime (days)
(C)	<u>x</u>	SE
34	3.675	0.099
30	3.225	0.025
26	3.325	0.053
22	5.725	0.058

Table 4-16. Reproductive rate of M. muscaedomesticae at different initial population densities when reared in spent house fly media with the nematode, Protorhabditis sp., at 30 $^{\pm}$ 2 C, 60-98% R.H.

	No. adult	female mi	tes introdu	uced (N _t)		
	10	20	40	80		
No. mites after 7	2 hrs (N _{t+}	L)				
\overline{X}	673.5	1243.0	1605.75	2663.0		
SE	29.6	283.4	65.2	84.2		
Reproductive rate per generation (R)*						
	66.35	61.15	39.14	27.23		
Timesa wearestan	11					

Linear regression line for R

$$R = 69.85 - 0.57 N_{+}$$

^{*} N_{t+1} - N_t

The study time (72 hrs) of this experiment was close to the generation time (3.225 days) of this mite species under experimental conditions (Table 4-15). Besides, these mites may multiply for a while in the Tullgren Berlese funnels when they are being separated from the rearing media. Therefore, the mean number of mites per box in each treatment was treated as N_{t+1} to calculate the reproductive rate per generation (R). The interception (69.85) of the linear regression of R was the extrapolated $R_{\rm O}$ (net reproductive rate) of $\underline{\rm M}.$ $\underline{\rm muscaedomesticae}$ under the studied conditions. The intrinsic rate of increase (r) was calculated by

$$r = \frac{\ln R_0}{T}$$

where T was the mean generation time (Birch 1948). The r of this mite species under the studied conditions was $\ln 69.85/3.225 = 1.3167$.

The non-linear equations, $N_{t+1} = N_t \exp(r(1-N_t/K))$ and $N_{t+1} = N_t (1+r(1-N_t/K))$, which were discussed by May (1974a, 1975), were used to predict the population growth in the propagating colony and to predict the harvest time. Neither of them described the actual data in mass-production studies. The calculated K value was too small (in hundreds) and a negative population resulted from the calculation.

The simplistic model was discarded as the intrinsic biology of this mite species was more fully

evaluated under laboratory and field conditions. The biological characteristics of this species include facultative larviparity, arrhenotoky, egg size (very small and hidden), and delicacy (sensitive to environment) of preimaginal stages. In preliminary studies on the life cycle of this mite species, a larva was seen 10 minutes after a previous observation during which there was no larva. This larva was apparently laid larviparously. Larviparity was induced later by the following technique: 1. Feed the adult female mites with frozen house fly eggs. 2. After the body of the female mites was swollen, a sign of a well-functioning reproductive system, let the environmental quality decline. The frozen fly eggs decomposed rapidly at 30 C. This would cause the female mites to retain their eggs without laying them. 3. Supply with fresh frozen fly eggs again. After step 2, the fully developed larvae were dissected from adult female mites by this researcher. The arrhenotoky exhibited by this mite species appears to be a major way of adjusting the sex ratio, probably through the "probability of insemination" as discussed by Filipponi et al. (1971, p. 210). This may also be one of the reasons for the differing sex ratio obtained by various authors (this researcher, Table 4-9, 4-12, Figure 4-2; Filipponi et al. 1971; Ito 1977b; Wade and Rodriguez 1961).

The Ro and r of this study were larger than the study of Filipponi et al. (1971) ($R_0 = 58.07$, r =0.9057). The mean generation time (T) of this mite species was 3.225, 4.48, and 5.117 days when fed on nematodes (this research), nematodes and frozen house fly eggs (Filipponi et al. 1971), and frozen house fly eggs (this research), respectively. In addition, these mites are phoretic and display what Butler and Cromrov have termed a "catastrophic behavior." This catastrophic behavior is a mass migration or movement at any time when environmental conditions become unsuitable for development. This phenomenon was noticed in field experiments described in Chapter 3. Farish compared the attractiveness of the manure and house fly to the mites. He found that when manure was more attractive than house flies, the mites remain in the manure, but when manure was less attractive than the flies, phoresy occurs (Axtell 1969). Phoretic behavior is common in the family Macrochelidae. With this species, attachment to either house flies or beetles is a common means of dispersal.

All these studies indicate that environmental conditions have an effect on the performance of this mite species. Figure 4-6 illustrated some of the known interactions determined in the course of this research that complicate model formation.

Previous research has indicated that there is a decline in the quality of rearing media after the 9th

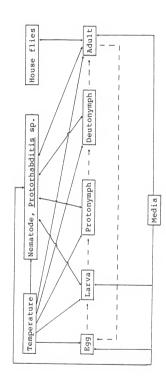


Figure 4-6. Diagramatic representation of biological and enviromental factors on life history of \underline{M}_{\bullet} . $\underline{muscaedomesticae}_{\bullet}$

day. This depletion of media included contamination resulting from the multiplication of bacteria that served as the food of nematodes and the contamination by ascid mites. The study on the reproductive rate of this mite species was conducted under 4 temperatures (the same as the study on generation time). Under temperatures other than 30 C, either fewer numbers of mites were produced or there were a larger proportion of immature stages—a reduced reproductive rate. The quality of the food and the population density influenced the reproductive rate and the developmental rate.

Although a number of the factors that influence and interact with the mite are now known (Figure 4-6), the difficulty is in assessing the effect of each factor on total production of the mite.

May (1974b) discussed the stability of population equilibrium in relation to a lagging density-dependent effect with lag-time T. T is not necessarily the same as the T (generation time) of this study. Nevertheless, if a lag effect of about a generation occurs, its effect on population dynamics will be about as follows. If rT is less than $\pi/2$, there is a stable equilibrium point at population K. If rT exceeds $\pi/2$, this potential equilibrium point is unstable, the population then oscillates, and the oscillation amplitude increases as rT increases. The rT for this mite species

under the studied conditions was ln 69.85 = 4.246, larger than π . If one computes the rT for 7 species of Macrocheles from the data of Cicolani (1979), the following results are obtained:

species	rT
M. matrius	3.67
M. robustulus	3.68
M. sabbadius	3.94
M. muscaedomesticae	4.06
M. perglaber	4.61
M. peniculatus	4.32
M. penicilliger	3.75

These data all indicate populations that will oscillate if lag effects of about one generation occur. The results may be due to media, nematodes, or any factor in which density at (-T) affects growth at present time. Therefore, it is reasonable to suspect that even if media were maintained somehow, large-amplitude oscillations might be observed.

In summary, the laboratory studies, although not directly applicable to field situations, indicate that the 8th day is the ideal harvest time at 30 C based on considerations of media depletion, nematode population, and the inherent nature of the oscillating population. It would also appear that the model of this mite species in mass production should include the nematode

population as a prey species—that is, it should be at least a two-species model.

CHAPTER 5

RELATIVE TOXICITY OF NINE INSECTICIDES AGAINST M. MUSCAEDOMESTICAE

Materials and Methods

The M. muscaedomesticae used in this study were from the stock colony described in Chapter 4 and had been maintained in the laboratory for over 8 months. The mites from the 7-day-old stock colony were separated from the rearing media by Berlese funnels into a glass jar. A piece of damp C-299 maiden chiffon of approximately 13 x 13 cm² was put in the jar to supply moisture and shelter. A layer of frozen house fly eggs was spread on the center of the cloth to provide food for the mite. The adult female M. muscaedomesticae in the glass jar were then tested within 2 days after their separation from the rearing media.

Those insecticides that are currently being registered for control of poultry pests in Florida (Koehler 1980) were selected for evaluation. These are carbaryl (1-naphthyl-N-methylcarbamate) coumaphos (O-(3-chloro-4-methyl-2-oxo-2H-1-benzopuran-7-yl) 0,0-diethyl phosphorothioate)

dichlorvos (0,0-dimethyl 2,2-dichlorovinyl phosphate)

malathion (diethyl mercaptosuccinate, S-ester with

0,0-dimethyl phosphorodithioate)

cyromazine (N-cyclopropyl-1,3,5-triazine-2,4,6-triamine)
These included 1 carbamate, 6 organophosphates, 1
synthetic pyrethroid, and 1 insect growth regulator.
The sources of these pesticides are given in Appendix 3.

The testing method described by Matthysse et al. (1975) was followed with only a slight modifications for M. muscaedomesticae. A 1% (weight per volume) stock solution of insecticide was formulated using Fisher brand reagent grade 99.8% methanol as the solvent for coumaphos and larvadex and the 99.5% acetone as the solvent for the others. Dilutions were made by either pipetting from these stocks or by serial dilutions using the same solvent as stock solution. Dilutions were geometric progressions, except for a very narrow series that were arithmetic progressions. The Fisher brand disposable borosilicate glass pasteur pipets, 14.6 cm in length, were used. The pipet was immersed in

the insecticide solution for 10 seconds to coat the interior surface. Then the pipet was emptied. The exterior deposit and the excess solution inside the pipet were removed with absorbent paper by touching the outside and both ends of the pipet. Blowing air through the pipet, as Matthysse et al. did, resulted in virtually no mortality. Therefore, the pipet was held vertically in a beaker for 30 minutes to dry the inside. The wide end was then covered with C-299 maiden chiffon and fastened with a rubber band.

Using an Emerson 1/4 HP vacuum/pressure pump, 20 female adult M. muscaedomesticae were drawn into each pipet from the glass jar. After collecting 20 mites, the pointed end of the pipet was sealed with Critoseal. The pipets containing the mites were then placed on acrylic plastic racks and kept at 21.1-29.4 C,50-70% R.H. Mortality was observed after 24 hours.

For the test procedure, each insecticide was replicated 4 times, i.e. 4 pipets, with 5-7 concentrations. Acetone— or methanol—treated pipets served as checks. Results from 2-3 tests were used to calculate LC50, LC95, and their confidence limits, as determined with use of the SAS computer program. When the toxicity to $\underline{\mathbf{M}}$. $\underline{\mathbf{muscaedomesticae}}$ was low, the insecticides were tested at a concentration as high as 1×10^{-2} .

Results and Discussion

Nine different pesticides are currently in use in Florida poultry houses to control flies and other noxious arthropod pests. Their application may possibly affect the population of M. muscaedomesticae. especially when applied onto poultry manure to control filth flies. The relative toxicity of these pesticides to M. muscaedomesticae is shown in Table 5-1. Six of these 9 pesticides have a very low toxicity to this mite. Four of them (dichlorvos, dimethoate, malathion, and stirofos) are registered for application in poultry houses at the concentration of 1×10^{-2} ai. Therefore. they were tested up to 1 x 10^{-2} . The mortality of M. muscaedomesticae that were treated with 1×10^{-2} of these 4 pesticides was 15.52%, 14.50%, 46.28%, and 28.78%, respectively. None of them killed 50% of the mites. Only malathion killed ca 50% of the mites. The other 2 (coumaphos and cyromazine) were applied in lower concentration. In addition, these 2 compounds had a low solubility in acetone, and precipitated as a 1 \times 10^{-2} solution in methanol. Therefore, these 2 pesticides were tested up to 5 x 10^{-3} . At this concentration, both coumaphos and cyromazine were virtually nontoxic to M. muscaedomesticae. Coumaphos is registered for application in poultry houses at a 1-2 \times 10-3 concentration to control lice or northern fowl mites. With this testing method, these concentrations

Table 5-1. Relative toxicity of 9 pesticides to the adult female \underline{M} , \underline{M} , \underline{M}

000+10100	0	Fiduci	Fiducial limit	1	Fiduci	Fiducial limit
		lower	upper	5627	lower	upper
coumaphos	1	1	1	ł	1	
cyromazine	}	1	7	1	1	1
dichlorvos	1	1	1	1	!	1
dimethoate	1	;	ł	}	1	1
malathion	1	;	;	;	1	!
stirofos	1	;	;	1	1	1
carbaryl	4.86x10 ⁻⁵	2.90x10 ⁻⁵	7.16x10 ⁻⁵	1.52×10 ⁻⁴	0.92×10-4	11.92×10-4
naled	1,68x10-3	1.60x10-3	1.77x10 ⁻³	2.49x10 ⁻³	2.27x10 ⁻³	2.90x10 ⁻³
permethrin	1.58×10-4	1.26x10-4	1.94×10-4	2.02×10-2	2.02x10-2 1.15x10-2	

Table 5-1. Continued.

- 1 - 1 - 1		Mortality highest co	Mortality (%) at the highest conc. tested
י בי	kegression equation	5×10-3	1x10-2
coumaphos	1	2.83	1
cyromazine	;	0.34	1
lichlorvos	!	1	15.52
dimethoate	;	1	14.50
nalathion	;	1	46.28
stirofos	1	;	28.78
carbaryl	Y=10.36+3.33X	;	1
naled	Y = 31.73 + 9.63X		
oermethrin	Y=7.97+0.78X	1	1

will not kill the macrochelid mites. Cyromazine was applied as a food additive in low concentration. It is an insect growth regulator and was expected to be non toxic to M. muscaedomesticae by this testing method. Three pesticides were toxic to this macrochelid mite with this testing technique. They were carbaryl, naled, and permethrin. The LC50 and LC95 for these compounds to this mite were 4.86 \times 10⁻⁵ and 1.52 \times 10⁻⁴, 1.68 \times 10^{-3} and 2.49 x 10^{-3} , and 1.58 x 10^{-4} and 2.02 x 10^{-2} , respectively. At a concentration of 1.5 x 10^{-4} . carbaryl, permethrin, and naled killed approximately 95%, 50%, and 1%, respectively, of adult female M. muscaedomesticae. Carbaryl is registered for application in poultry houses at 45×10^{-4} concentration to control lice, fowl tick, and northern fowl mite. At this concentration, it has the potential for causing 100% mortalitly of macrochelid mites. However, since it would not be applied directly to the poultry manure but to the birds, its effect on the macrochelid mite population will be somewhat reduced. Permethrin is registered for application in poultry houses at a concentration of 1.12×10^{-3} to control the adult house fly. At this concentration, it could produce 75% mortality of the macrochelid mite. Again, it is not applied directly to the manure. Naled also is registered for application in poultry houses at a concentration of approximately $2-3 \times 10^{-3}$, a

concentration that has the potential to kill 95% of the macrochelid mites; but again it is not normally applied to the manure.

At the LC₅₀ level, carbaryl was most toxic to the mites, followed by the permethrin and then the naled. Carbaryl was approximately 3 times more toxic than permethrin and 34 times more toxic than naled to M. muscaedomesticae. At the LCos level, carbaryl was still the most toxic to these mites. But naled became more toxic than permethrin at this level. At this level. carbaryl was 16 times more toxic than naled and 133 times more toxic than permethrin. In comparing the regression lines, the slope of naled is very steep (9.63), the slope of carbaryl is flattened (3.33), and the slope of permethrin is very flattened (0.78). The laboratory population of this mite is homogeneous to naled, but heterogeneous to both carbaryl and permethrin. Axtell (1966) studied the toxicity of 17 insecticides to the adult female M. muscaedomesticae in the laboratory. Coumaphos, dichlorvos, dimethoate, malathion, and naled were among the 17 insecticides studied. The method he used to determine the toxicity differed from this study. The insecticide solutions were mixed with CSMA house fly medium. The mites were then introduced into the medium for 24 hours. The $LC_{50}s$ of the above 5 insecticides to adult females of this mite species were 4.4 x 10^{-4} , 2.6 x 10^{-6} , 2.6 x

10⁻⁵, 2.8 x 10⁻⁵, and 4.4 x 10⁻⁴, respectively. The slopes of the regression lines were 0.697, 3.987, 2.390, 2.040, and 3.677, respectively, i.e., the mite population in his study was heterogeneous to all 5 insecticides. Though the results of these 2 studies were not comparable because of the different testing methods, it appears that in Florida this mite has developed resistance to these 5 insecticides. The flattened slopes of the regression lines of carbaryl and permethrin also indicated that resistant individuals existed in the mite population studied.

The short life cycle and high reproductive rate of Musca domestica are currently understood to be the 2 key biological characters for the development of pesticide resistance, and therefore, M. muscaedomesticae, which also has these 2 key characters, has a high potential for developing pesticide resistance. The poultry farm from which the laboratory colony of M. muscaedomesticae was started had been applying dichlorvos (Vapona 23.4% E.C.) for 7 to 8 years, and recently, a mixture of dichlorvos and stirofos to control the house fly. The pesticides were applied regularly twice a week in the fly season. Therefore, it is highly possible that this strain of mite was resistant to dichlorvos and stirofos prior to being brought into the laboratory. The resistance to coumaphos, dimethoate, and malathion has probably

resulted from either a cross resistance or the immigration of resistant individuals (genes) from other populations. As previously stated, cyromazine is an insect growth inhibitor. It appears to be selective toward dipterous species. The mechanism of toxicity of cyromazine is uncertain. It does not seem to act by inhibiting chitin synthesis as does diflubenzuron (Miller et al. 1981). Axtell and Edward (1983) studied the effect of cyromazine (Larvadex) as feed additives to the house fly and non-target arthropods in poultry manure. The house fly was effectively controlled, and there were no adverse effects on the population of manure-inhabiting mites (M. muscaedomesticae and Fuscuropoda vegetans) and histerid beetles (Carcinops pumilio). Yet, their data showed the population of the 2 mite species in the treated area increased when the mite populations in the untreated area decreased. This suggested cyromazine may have indirectly stimulated the population of mites to increase, instead of being toxic. Further studies of this problem may provide a selective chemical for the biocontrol of house flies.

Based on this study, coumaphos, dichlorvos, dimethoate, malathion, and stirofos preferably should be applied to control poultry pests because of their low toxicity to M. muscaedomesticae. When applying carbaryl, naled, and permethrin, care should be

exercised to avoid applying them to the manure so as to decrease their harmful effects on these mites.

At the present time natural enemies cannot by themselves hold fly population density below the economic or esthetic nuisance levels in all seasons. Insecticides have to be applied periodically at most poultry farms. After the application of insecticides, the house fly population resurges rapidly but the mite population increases slowly (Axtell 1963b, 1968; Wicht and Rodriguez 1970). This can cause great difficulties in an IPM program for house flies. In addition, the ability of the house fly to develop resistance to insecticides has been well documented. House flies have been reported to be resistant to organochlorines. organophosphates, carbamates, pyrethroids, juvenoids, and diflubenzuron, including coumaphos, dichlorvos, diazinon, malathion, naled, permethrin, and trichlorfon, which were used on poultry farms (Bailey et al. 1971, Georghiou et al. 1967, Keiding 1977, LaBrecque et al. 1958, MacDonald et al. 1983). Harris et al. (1982) reported the multiple resistance of the house fly to 7 organochlorines, 11 organophosphates, 5 carbamates, 8 pyrethroids, and pyrethrin. The resistance to permethrin was developed in one year. These pesticides included 7 of the pesticides studied in this current research, except for cyromazine and coumaphos. Consquently, the development of a pesticide

resistant strain of natural enemies to be incorporated into an IPM program for house fly control should be one of the principal and more immediate objectives in the future. Several predaceous phytoseiid mites have been reported to be resistant to pesticides in laboratory tests (Georghiou and Taylor 1976). A multipesticide-resistant strain of the predaceous phytoseiid mite, Metaseiulus occidentalis, was artificially selected and propagated in the laboratory, then released and subsequently established in the field (Hoy 1979; Hoy and Knop 1979; Hoy et al. 1980,1982; Roush and Hoy 1981). M. muscaedomesticae has as great a potential for future use as Metaseiulus occidentalis, since it already has shown resistance to several organophosphates. The artificial selection of a multipesticide-resistant strain of this mite would be relatively easy. As Anderson (1983) stated, the incorporation of pesticide-resistant predaceous mites into the IPM programs for house fly suppression would have many advantages. The pesticide-resistant strain would make it feasible to control pest populations but not eliminate the predators. Therefore, a better IPM program for fly control could be developed.

In addition to controlling the house fly, this mite could be the source material for the genetic study of pesticide resistance.

Parasitoid wasps are another group of important natural enemies of house fly. They appeared promising for the control of the house fly. Their incorporation into the IPM program of house fly management is a matter of course. The selection of a pesticideresistant strain of parasitoid wasps and/or the selection of selective pesticides are additional important objectives. They should be done to protect the benefits gained from the study of pesticide-resistant mites.

CHAPTER 6

THE EVALUATION OF THE CONTROL EFFECT OF $\underline{\mathsf{M}}_{\bullet}$ MUSCAEDOMESTICAE ON HOUSE FLY

Materials and Methods

Laboratory Test

The objective of laboratory testing was to determine the best ratio of mites to house fly eggs (M/E) to be used in the field test.

The standard substrate (ca 75 gm CSMA media plus 130 ml water per pan) was placed in aluminum pans which were 20 x 13.5 x 4.7 cm³ and left overnight to ferment before use. Eighteen hours later, 1000 house fly eggs were added to each pan. The house fly eggs were measured volumetrically, i.e., 0.1 ml contains approximately 1000 eggs. A thin layer of substrate was spread over the house fly eggs to facilitate egg hatching. Two hundred, 100, and 50 adult female M. muscaedomesticae from the laboratory propagating colony (see Chapter 4) were added to each pan together with the house fly eggs, i.e., in a M/E ratio of 1/5, 1/10, and 1/20, respectively. These mites were separated from the breeding colony one day before so that they could be inoculated in a short time. Pans without mites

served as checks. The house fly pupae in each pan were floated and counted 8 days after the addition of fly eggs and mites. Five pans were used for each treatment.

Field Test

Field tests were conducted in the range houses of the Department of Poultry Science, University of Florida. On either side of the house, a row of 15 cages was suspended 1 m above the ground, each containing one hen. Two houses were used, one for the release, the other as a check. The test was conducted in August and September (1984) when house flies are normally present in high numbers. The substrate--manure and sawdust under the cages--were removed to eliminate the soldier fly larvae, which would make the caged-hen manure unsuitable for the development of house fly larvae, and a sheet of plastic was placed under each row of cages. Then a thin layer of fresh caged-hen manure, removed from another poultry house, was placed on the plastic sheet for house flies to oviposit. Amdro 0.88% bait (Tetrahydro-5,5-dimethyl-2(1H)-pyrimidionoine(3-(4-(trifluoromethy1)pheny1)-1-(2-(4-trifluoromethy1)pheny11) ethenyl)-propenylidene)hydrazone) was applied to control the fire ants and other predatory ants. M. muscaedomesticae were released 3 times at 7-day intervals by depositing the propagating colony over the manure. The population of house fly larvae were sampled the day before the

release of the mites, i.e., at 7-day intervals also, to determine the number of mites to be released. This sampling was continued until the termination of this test. The area of manure with fly larvae activity was estimated. Manure samples of 1-2 cm deep and 40.5 cm² in area were taken with a flat shovel that was especially made for the sampling. Six manure samples, 3 under each row of cages, were taken from each range house. Each manure sample was placed into a 95% isopropanol in a glass jar to kill the arthropods. The number of 3rd-instar house fly larvae and adult female macrochelid mites were counted. The population of the 3rd-instar house fly larvae in each range house was estimated by

mean no. 3rd-instar house
fly larvae per sample = total area of manure with fly
larvae activity (cm²)

The method of Morgan et al. (1981) was then followed to estimate the amount of house fly eggs in the range houses, using a growth rate of 1 \times .

M. muscaedomesticae were released in a M/E ratio of 1/5. Each box was assumed to contain 2500 mites. The proportion of adult female mites in the box was 0.53 (Table 4-9), so that 1325 adult female mites were assumed to be in each box. This mite was released in box units. The estimated number of mites to be released was divided by 1325 and rounded to an integer; this

integer determined the number of boxes of propagating colony to be released.

Results and Discussion

Laboratory Test

The control effect of adult female \underline{M} . <u>muscaedomesticae</u> on the house flies is shown in Table 6-1. The mortality of the house fly (based on the reduction in the number of recovered pupae) was 99.7%, 92.3%, and 77.7%, respectively, when the M/E ratio was 1/5, 1/10, and 1/20. The 2 higher ratios, 1/5 and 1/10, gave significantly better control of the house fly than the ratio of 1/20.

Similar studies have been conducted by many researchers (Axtell 1963b, Filipponi 1955, Ito 1973, King 1964, O'Donnell and Axtell 1965, Peck 1969, Rodriguez et al. 1970, Singh et al. 1966). Most of their results agreed with this study. Mites added in the M/E ratio of 1/5 or 1/10 always gave good control of the house fly. The ratio of 1/20 resulted in lower mortality of the house fly. However, Axtell (1963b) added M. muscaedomesticae in a M/E ratio of 1/150 and still got a control of 97%. The study of Filipponi (1955) allowed the mites contact with the house fly eggs for only 2-9 hours. Therefore, the percentage of control in his study was low and would be higher if the

Table 6-1. Control of house fly by adult female $\underline{\text{M}}$. $\underline{\text{muscae-domesticae}}$ in CSMA medium with varying ratios of $\underline{\text{mite}}$ to house fly egg.

	Ratio of mite/house fly egg			
	CK	1/5	1/10	1/20
No. eggs used	1000	1000	1000	. 1000
No. mites used	0	200	100	50
No. pupaee recovered				
X*	459.6c	1.2a	35.6a	102.6b
SE	20.5	0.7	9.0	34.5
Percentage of control		99.7	92.3	77.7

^{*} Mean of 5 pans. Means followed by the same letter were not significantly different at the 0.05 level in Duncan's multiple range test.

mites were allowed to stay with the fly eggs. The study of Ito (1973) showed that in the presence of other foods, the control effect of these mites on house fly was reduced. A previous study had added naeatode medium together with house fly eggs and mites to CSMA medium. Its results are shown in Table 6-2. The percentage of control was lower than that shown in Table 6-1 in each treatment. The M/E ratio of 1/5 was chosen for the field test, because there is abundant additional food in the poultry manure.

Field Test

Five days after the removal of old manure and sawdust that contained mainly soldier fly larvae (the day of "-1"), sampling of the manure showed a house fly population established. Three consecutive releases of M. muscaedomesticae were done at 7-day intervals. The results of these tests are shown in Table 6-3. Manure samples were taken 3 times, each, 1 day before the release of the mites. The population size of the 3rd-instar house fly larvae in each range house was estimated from these manure samples. The growth rate of the house fly population was assumed to be 1 x. Following the numbers in Table 3 of Morgan et al. (1981), the population size of house fly eggs in the range house was estimated from the population size of the 3rd-instar house fly larvae. Then M.

Tabel 6-2. Control of house fly by adult female $\underline{\text{M}}$. $\underline{\text{muscae-domesticae}}$ in CSMA medium with different ratios of mite/fly egg when nematodes were added.

	Ratio	of mite/h	ouse fly	egg
~ ~ ~ ~ ~ ~ ~ ~ ~ ~ ~ ~ ~ ~ ~ ~ ~ ~ ~ ~	CK	1/5	1/10	1/20
No. eggs used	2700	2700	2700	2700
No. mites used	0	540	270	135
No. pupae recovered				
<u>x</u> *	2423.6c	302.8a	974.0b	1225.0b
SE	35.3	81.4	102.4	164.9
percentage of control		88.0	61.3	51.3

^{*} See Table 6.1.

muscaedomesticae were released at a ratio of 1/5 to house fly eggs. Twelve, 16, and 6 boxes of mites were released at days 0, 7, and 14, respectively. Before the first release of the mites, both range houses had approximately the same house fly population. By the 6th day, the house fly population increased in both houses. However, the treated house had a smaller house fly population than the check house. During the remainder of the test, the house fly larvae were more abundant in the treated house than in the check house. The test was terminated after 23 days when the hens were removed.

Generally, this test had a negative result.

However, the house fly population at the 6th day did show that this mite exhibited some control on field house fly populations. Several factors caused the negative results in this test.

- The spent house fly media that is deposited together with the mites is an excellent medium for the breeding of house flies.
- 2. There was vigorous activity of fire ants in the range house.
- 3. There was recovery of soldier fly larvae.
- 4. The leakage of drinking water increased the moisture content of the manure.
- 5. $\underline{\text{M}}$. $\underline{\text{muscaedomesticae}}$ was not only present in the check house, but its population multiplied.
- 6. There was migration of adult house flies.

7. There was a lack of a "manure base" to absorb moisture from the freshly deposited manure.

Although this field test had a negative result, it is the belief of this researcher that the augmentative release of these mites to control the house fly is feasible, based on the research described in Chapter 3 and on the laboratory tests reported in this chapter. The poor results of the field test indicate again that the mites are unable to control the house fly during the summer fly season. Therefore, supplemental control is necessary, which would require additional studies.

Applying the method of Morgan et al. (1981) to the data of field samples in the study of population fluctuation in Chapter 3 of this research, the estimated fly egg population exceeded 5 x that of the field adult female M. muscaedomesticae population on 2 occasions. The first time was on November 4, 1983, and then on June 5, 1984. Both dates were at the time when the population of fly larvae was peaking. On June 29, 1984, the estimated fly egg population was approximately 5 x that of the field population of adult females of this mite species. After this, the larval population dropped, and the estimated fly egg population was lower than 5 x that of the adult female mite population. Currently, this may be the only scouting method for IPM program to determine the time to release parasitoid wasps and/or these mites-whenever

field sampling shows that the population of this mite is too low (\leq 1/5 of estimated fly egg population).

CHAPTER 7

CONCLUSION

There are several major conclusions that have been arrived at as a result of this research. In addition, based on results obtained in both the laboratory and the field, a suggested IPM program is proposed for control of house flies on poultry farms in Florida as well as for future research to be considered.

The conclusions drawn from this research are

The predaceous macrochelid mite, M.

muscaedomesticae (Scopoli), is the key predator in suppression of the population of the house fly on poultry farms in Florida. This is based on: a. The negative density dependence between the field population of this mite species and the house fly.

b. Considering those predators that inhabit poultry manure, only this mite species has the same distribution as the house fly in the different moisture levels of the poultry manure. The other predators distribute themselves in drier manure levels and consequently are away from the majority of the house fly population.

The research on mass production revealed the requirement of a rhabditid nematode, <u>Protorhabditis</u> sp., in the diet of this mite.

This mite species is an excellent candidate for the biocontrol of the house fly. This is based on: a. The number of house fly eggs consumed per mite per day. b. The high probability of artificially selecting pesticide-resistant strains of this mite species.

- c. The ability to mass produce these mites cheaply.
- d. The 99.7% control of house flies obtained in laboratory tests.

This research has indicated that when any of the following events occur, $\underline{\mathsf{M}}.$ $\underline{\mathsf{muscaedomesticae}}$ should be released if control is to be maintained.

- Immediately after manure removal, since most of the mites will be removed with the manure. The remaining manure base is dry and contains very few of the mites.
- After drastic temperature changes, which are disastrous to the mite population.
- During the summer fly season. However, the release of parasitoid wasps may be a better choice at this time.
- After the application of larvicide to control flies.
 The pesticide-resistant mite strains will be invaluable.
- 5. When the scouting method suggested in Chapter 6 shows the necessity.

It should be noted that when mites are released, the rearing media should not be released or should be isolated from the house flies, e.g., placed in a screen cage. This media is ideal for house flies to lay eggs.

The integrated control techniques suggested by Axtell (1968, 1969, 1970a,b, 1981), Krantz (1983), Peck and Anderson (1970), Rodriguez et al. (1970), Singh and Rodriguez (1969) and Wicht and Rodriguez (1970) should also be followed. These techniques are

- Apply adulticide preferentially, spot-spraying larvicide.
- Use selective formulations of pesticide, e.g., bait, trap.
- Remove manure after a long interval and early before the fly season so that the population of natural enemies can recover.

However, a good manure management strategy is the basis for a successful IPM program to control the house fly.

Further studies should be conducted on the following problems.

- 1. The reasons for the failure of \underline{M} . \underline{M} muscaedomesticae to control house flies during the summer fly season in Florida. Study on this problem should have the first priority.
- 2. The influence on the population of $\underline{\texttt{M}}$. $\underline{\texttt{muscaedomesticae}} \ \ \text{by different manure management}$

strategies, e.g., rotovation. Then various IPM programs can be developed for the different manure-management strategies.

- 3. The combined effect of $\underline{\mathsf{M}}$. $\underline{\mathsf{muscaedomesticae}}$ and parasitoid wasps in controlling house flies.
- 4. The improvement of the mass-production method.
- 5. The selection of pesticide-resistant strains with both this mite species and parasitoid wasps.
- 6. The effect of poultry molt on the manure and manure-inhabiting arthropods.
- An economic threshold that should be standardized.
 The following problems may not be as important as

the former ones, but also should be studied.

- 1. The effects of other manure-inhabiting predators.
- 2. The possibility of using other macrochelid species to control house flies.
- 3. A method to mass produce rhabditid nematodes and spray them on the poultry manure to see if this can induce an "outbreak" of macrochelid mites and, consquently, reduce the house fly population.

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APPENDIX 1

THE FORMULA OF HOUSE FLY MEDIUM

Brewers dried grain (Howland Feed Co.),	40%.
Soft wheat bran (Brownlee Feed and Seed, Co.),	33%.
Pelletized coastal Bermuda (Whistling Pines Ranch),	27%.
Water, 4-5 li	ters.

APPENDIX 2

THE PLASTIC BOXES (32- AND 80-OUNCE)
AND THE MEATBALLER



Figure A-1. The plastic boxes (32- and 80-ounce) and the meatballer. $\,$

APPENDIX 3

SOURCE OF PESTICIDES STUDIED IN CHAPTER 5

Carbaryl Union Carbide Agricultural Products

Company, Incorporated

Coumaphos Mobay Chemical Corporation

Cyromazine CIBA-GEIGY Corporation

Dichlorvos Shell Development Company

Dimethoate American Cyanamid Company

Malathion American Cyanamid Company

Naled Ortho Division, Chevron Chemical

Company

Permethrin ICI Americas, Incorporated

Stirofos Ortho Division, Chevron Chemical

Company

BIOGRAPHICAL SKETCH

Mr. Chyi-Chen Ho was born on August 23, 1950, in Taipei, Taiwan, Republic of China. He graduated from Hsing-Chu high school, Hsingchu, Taiwan, in 1967. In 1971, he received a Bachelor of Science degree in entomology from the National Chung-Hsing University, Taichung, Taiwan. He then served in the army of the Republic of China for 2 years. In 1973, he enrolled in graduate school in National Taiwan University, Taipei, Taiwan. He received the Master of Science degree in entomology in 1975.

From 1975 to 1976, he was employed as an entomologist for Taiwan Banana Research Institute, Pintung, Taiwan. He changed jobs in 1976 to the Taiwan Agricultural Research Institute. He was granted a scholarship from his country in 1980 for advanced studies. From 1931 to present, he has been a graduate student in the Department of Entomology and Nematology, University of Florida, studing toward a degree of Doctor of Philosophy.

Mr. Ho is married to Su-In Tsai and has two sons. He is a proud, happy husband and father.

I certify that I have read this study and that in my opinion it conforms to acceptable standards of scholarly presentation and is fully adequate, in scope and quality, as a dissertation for the degree of Doctor of Philosophy.

Henry L. Nonnoy

Harvey L. Cromroy, Chairman Professor of Entomology and Nematology

I certify that I have read this study and that in my opinion it conforms to acceptable standards of scholarly presentation and is fully adequate, in scope and quality, as a dissertation for the degree of Doctor of Philosophy.

Achard S. Patterson

Professor of Entomology and

Nematology

I certify that I have read this study and that in my opinion it conforms to acceptable standards of scholarly presentation and is fully adequate, in scope and quality, as a dissertation for the degree of Doctor of Philosophy.

Philip G. Koehler

Professor of Entomology and Nematology This dissertation was submitted to the Graduate Faculty of the College of Agriculture and to the Graduate School, and was accepted as partial fulfillment of the requirements for the degree of Doctor of Philosophy.

May 1985

Dean, follege of Agriculture

Dean for Graduate Studies and Research

I certify that I have read this study and that in my opinion it conforms to acceptable standards of scholarly presentation and is fully adequate, in scope and quality, as a dissertation for the degree of Doctor of Philosophy.

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